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UKTAG Environmental Quality Standards

Recommendation for Emamectin Benzoate

by

Water Framework Directive – United Kingdom Technical Advisory Group (WFD-UKTAG)

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Executive Summary

The purpose of this report is to derive Environmental Quality Standards (EQS) for the substance emamectin benzoate (EMB; CAS number 155569-91-8). Emamectin benzoate is an insecticide which is currently only approved for use in the UK as a veterinary medicine for the treatment of sealice in farmed Atlantic salmon. Environmental Quality Standards can be derived for different environmental compartments, ie water, sediment and biota, depending on the properties of the substance. For EMB, an EQS for sediment and EQS for water (protective of pelagic organisms) in relation to long term and short term exposures have been derived according to technical EQS guidance (EU, 2018) produced under the Water Framework Directive (EU 2000). Biota standards were not derived as, based on the EQS guidance and the properties of the substance, they were not identified as required for EMB. Due to the current use of EMB, derivation of an EQS for the marine environment only has been undertaken (should the substance's use pattern substantially change in the future, this could be revisited). A Quality Standard (QS) for drinking water was not necessary because only the marine environment is being considered.

The Chemistry Task Team (CTT) of the UK's Technical Advisory Group (UKTAG) previously produced a draft EQS report for the substance in 2019 which was publicly consulted on. Following the consultation responses and the availability of new reliable and relevant laboratory ecotoxicty and field monitoring data, the report has been revised and subsequently peer reviewed. This final report therefore represents new data available since the consultation, comments received during the consultation, and comments arising from the peer review.

In producing this report key information sources considered include: a peer reviewed EQS report commissioned by the Scottish Environment Protection Agency in 2017; three field studies reported in 2018 and 2019 (and a statistical reanalysis of two of these); results of older and more recent ecotoxicity testing conducted by industry and the results of a literature search to update the database of relevant toxicity data for EMB.

The table below summarises the proposed EQS with a brief summary of the basis for their derivation.

$EQS_{sediment, sw\ eco}$ (ng/kg dwt)	MAC-$EQS_{sw, eco}$ (ng/L)	AA-$EQS_{sw, eco}$ (ng/L)
272	1.2	0.17
Based on the lowest relevant & reliable study (28-day *Chironomus riparius* NOEC 2720 ng/kg dwt, normalised to default 5% organic carbon content). An assessment factor of 10 was applied; chronic data was available for 2 insect species, 1 polychaete species and 4 crustacean species. Species differences in living/feeding strategies supported the use of an assessment factor of 10. Field data provides additional confidence the assessment factor is appropriate and the derived EQS is protective.	Based on mean of two lowest relevant & reliable studies (96-hour *Americamysis bahia* LC_{50}s 78 & 40 ng/L). Assessment factor of 50 applied; data available for 2 primary producers, 8 crustacean species, 1 marine mollusc, 2 insect species and 4 fish species. Lacking details for an additional lobster study and the potential for additional exposure via ingestion justify the use of the assessment factor of 50 rather than a value of 10.	Lowest relevant & reliable study (28d *Americamysis bahia* NOEC (growth) 8.7 ng/L). Assessment factor of 50 applied; data available for 1 primary producer, 3 crustacean species and 1 fish species. Differences in living/feeding strategies were considered insufficient to enable use of a lower assessment factor of 10.

dwt – dry weight

AA – Annual Average

MAC – Maximum Allowable Concentration

$EQS_{sw,eco}$ – Environmental Quality Standard (EQS) for saltwater (SW)

The AA-$EQS_{sw, eco}$ is very similar to that proposed in 2019 (0.17 cf 0.2 ng/L). The difference reflects a slightly lower available test result being used for the derivation. The MAC-$QS_{sw, eco}$ proposed here, 1.2 ng/l, is lower than that proposed in 2019 (7.8 ng/l), mainly because a larger assessment factor has been used following consideration of consultation comments. The $EQS_{sediment, sw\ eco}$ (272 ng/kg dwt) is about 11.5 times less stringent than that proposed in 2019 (23.5 ng/kg dwt). This has resulted from a five-fold reduction in the applied assessment factor, reflecting the much increased database of chronic sediment studies that is now available, and using a normalised metric of a default sediment organic carbon content of 5%.

Glossary

AA	Annual Average
ADI	Acceptable Daily Intake
AZE	Allowable Zone of Effect
BCF	Bioconcentration factor
BioSS	Biomathematics and Statistics Scotland
CAS (number)	Chemical Abstract Service (number)
CCA	Canonical Correspondence Analysis
CIS	Common Implementation Strategy
CRED	Criteria for Reporting and Evaluating Ecotoxicity Data
CTT	Chemistry Task Team of the UK's Technical Advisory Group
DT_{50}	Half life for dissipation
Dwt	dry weight
EC	European Commission
EC_{10}	Concentration at which a specified effect endpoint is observed in 10% of the test population
EC_{50}	Concentration at which a specified effect endpoint is observed in 50% of the test population
ECO	Ecological
EFSA	European Food Standards Authority
EMB	Emamectin benzoate
EPA	US Environment Protection Agency
EQS	Environmental Quality Standard
EU	European Union
GABA (receptors)	*gamma* aminobutyric acid (receptors)
GAMM	Generalised Additive Mixed Model
GLMM	Generalised Linear Mixed Model
HSE	Health and Safety Executive
K_{oc}	Partition coefficient between water and organic carbon
K_{ow}	Partition coefficient between octanol and water
LC_{50}	Concentration at which mortality is observed in 50% of the test population
LMM	Linear Mixed Model
MAC	Maximum Allowable Concentration
NOAEL	No observable adverse effect level
NOEC	No Observable Effect Concentration
OC	Organic Carbon
OECD	Organisation for Economic Cooperation and Development
QS	Quality Standard
SAMS	Scottish Association for Marine Science
SEPA	Scottish Environment Protection Agency
SW	Salt Water
TOC	Total Organic Carbon
UKTAG	UK's Technical Advisory Group for the WFD
WFD	The Water Framework Directive
Wwt	Wet weight

1. Introduction

1.1 Background to the report

The Chemistry Task Team (CTT) of the UK's Technical Advisory Group (UKTAG) was asked in 2018 to recommend an environmental quality standard (EQS) for the fish farm sealice medicine emamectin benzoate (EMB). The Scottish Environment Protection Agency (SEPA) previously derived standards for the substance in 1999, before current EQS development guidance and significant new data were available. In 2019 a draft UKTAG EQS report for EMB was produced by CTT and consulted on. A significant number of comments were received along with subsequent submission of new ecotoxicological data and notification of ongoing field study work (see Annex 2). In response UKTAG committed to reviewing the EQS proposal based on consideration of the comments received and the new toxicity data and field study information. This report derives revised EQS for EMB and addresses the points raised in the previous consultation and a subsequent peer review (see Annex 4).

1.2 Standards Considered

Environmental Quality Standards (EQS) can be derived for a number of different endpoints (EU, 2018). These include the derivation of Quality Standards (QS) for the water column to protect aquatic life, derivation of a QS for sediment to protect sediment organisms, and derivation of a QS for biota, either in relation to secondary poisoning or the protection of human health from the consumption of fish. In addition, a QS in relation to drinking water can also be derived for application in those waters where abstraction for drinking water occurs.

Quality Standards for the water column are derived for all substances, with QS usually derived for freshwater and saltwater in relation to effects arising from short term exposure and long term exposure. The QS derived for short term exposure is the Maximum Allowable Concentration (MAC) and is designed to be protective of short term, intermittent exposures. The QS for long term exposure is the Annual Average (AA) which is designed to be protective of longer term, sublethal impacts. Quality Standards for sediment and biota are derived if the behaviour and hazardous properties of the substance mean that such QS values are relevant. For example, a QS for sediment is derived where the data for a substance indicates it is likely to adsorb to sediment (eg it has a log Koc of >3) and the derivation of a biota QS is dependent on the properties of the substance in relation to bioaccumulation and toxicity to mammals.

In this report QS have been derived for EMB for surface waters (ie a Maximum Allowable Concentration, MAC-$QS_{sw,\ eco}$, and Annual Average, AA-$QS_{sw,\ eco}$). Based on the properties of the substance (see section 2.3), a QS for sediment, $QS_{sediment,\ sw\ eco}$, has also been derived. Due to the substance's current use pattern in the UK, standards in water and sediment have only been derived in relation to the marine environment (saltwater). Standards for secondary poisoning and humans exposed via the environment have not been derived as the substance does not meet the criteria for this assessment (eg EMB's measured bioconcentration factor is less than 100 l/kg). Neither has a standard for surface water for drinking water abstraction been derived based on the substance's use pattern (in the marine environment). Should the substance's use pattern significantly change in the future, this can be revisited.

1.3 Data Sources and Methodology

The proposed QS for EMB have been derived using the European Union's Common Implementation Strategy (CIS) Technical Guidance number 27, "Technical Guidance for Deriving Environmental Quality Standards" (EU, 2018), here referred to as the EQS technical guidance. This guidance has been used to derive EQS that are currently in place as statutory EQS in the UK and is the current guidance in the UK for EQS derivation. As noted above (Section 1.2) however, QS have only been derived for the

saltwater environment and not freshwater, as based on the current use of EMB (Section 2.1) the current focus is on the marine environment.

Data used in this assessment came from three main sources: a 2017 EQS proposal report commissioned by SEPA (2017), additional studies made available or commissioned by industry after 2017 and more recent studies retrieved from the academic literature. These are described briefly below.

The 2017 EQS proposal report (SEPA, 2017) reviewed additional ecotoxicity data that had become available since the previous standards were set by SEPA in 1999 and proposed EQS based on the available guidance, which has since been updated (EU 2018). The 2017 report identified the key QS for EMB as sediment and the water column.

New test data available since the 2019 public consultation, are described in section 3.2.1. Studies identified in the academic literature through key word searches using ScienceDirect and SCOPUS, include a more recent microcosm experiment (section 3.2.2) and further laboratory fate and ecotoxicity studies. Available field study data, including recent reanalysis, are described in section 3.2.3. Following the 2019 consultation UKTAG said that a statistical reanalysis of the data would be undertaken. Based on the reanalysis undertaken on behalf of the industry and described here, this was no longer seen as necessary.

The relevance and reliability of data used in the derivation of the QS for EMB have been appraised following the principles of the Klimisch code approach and CRED systems (Klimisch *et al*, 1997; Moermond *et al*, 2015). However, for the data contained in the EQS proposal report (SEPA, 2017) only selected key studies have been further reviewed here. This approach is justified in that consultants engaged for that work carried out a review of reliability according to Klimisch *et al* (1997), and many of the studies have been reviewed and used in recognised EU and international regulatory programmes.

2 Information on the Substance

2.1 Uses

The only known current authorised use of emamectin benzoate in the UK is as an in-feed medicine in finfish aquaculture to control sealice, e.g. *Lepeophtheirus salmonis*, in salmonids (Veterinary Medicines Directorate, 2022). It is approved for use in the EU as a plant protection product active (EU Pesticide Database) but there are no such products, containing EMB as an active ingredient, approved for use in the UK (HSE Pesticide Register). Use at Scottish fish farms per unit biomass and application has on average increased from around 26 µg/kg biomass/year in 2002 to a peak of 67 µg/kg biomass/year in 2015. There also has been an increase of average application rates from around 1.4 applications/site/year in 2002 to 2.68 applications/site/year in 2016 (SEPA 2017). It is of note that between 2002 and 2015 the amount of biomass in Scottish fish farms has doubled whereas the total mass of emamectin benzoate used in Scottish fish farms has increased six fold over the same period (SEPA 2017).

2.2 Substance identification

Emamectin benzoate ((4''R)-4''-deoxy-4''-(methylamino)avermectin B1 benzoate (CAS Number: 155569-91-8 (formerly CAS: 13751274-4 and CAS: 179607-18-2)) is a mixture of emamectin B1a (90%) and emamectin B1b (10%) (SEPA, 2017). Both isomers are large molecules (ca. 1000 relative molecular mass) and differ by one methyl group (EFSA, 2012) (figure 1).

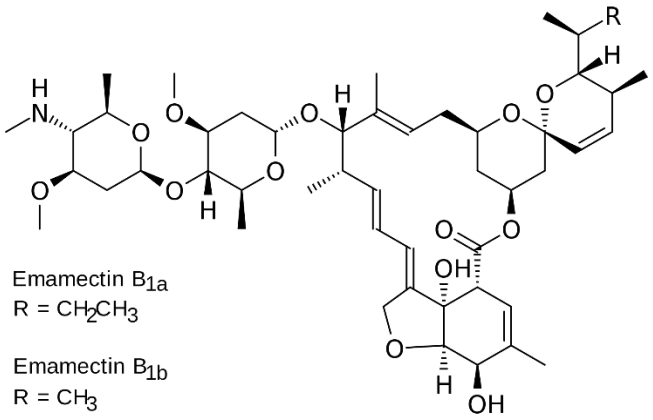

Figure 1 Molecular structure of emamectin (benzoate moiety not shown) (EFSA, 2012)

2.3 Physico-chemical properties

The substance is a white solid at 25 °C with a melting point of 160 °C and a low vapour pressure (4 x 10-6 Pa at 21°C) (EFSA, 2012). It has a water solubility of 24 mg/l at pH 7 (5.5mg/l in saltwater; OECD 105 flask method) (SEPA, 2017). The substance contains two functional groups that are ionisable at environmentally relevant pH (pKa's of 4.2 (benzoic acid) and 7.7 (epi-methyl-$NH2^+$; OECD 112 potentiometric titration) (SEPA, 2017). Consequently, the substance's octanol-water partition coefficient (log K_{ow}) varies between 3 and 5.9 in the pH range 5 – 9, with a log K_{ow} of 5 at pH 7 (OECD 107 shake flask method) (SEPA, 2017 and EFSA, 2012). Experimentally derived soil organic carbon-water partitioning coefficients (K_{oc}) were noted for a number of different soil types including (log K_{oc} in parentheses): 278,983 (5.45; sandy loam), 25,363 (4.40; clay loam), 28,325 (4.45; silt loam) (SEPA, 2017; EFSA, 2012).

2.4 Fate and behaviour

Emamectin benzoate is hydrolytically stable between pH 5.2 and 8 but at pH 9 a DT_{50} of 19.5 days was reported at 20 °C (EFSA, 2012). Several studies on photolysis of EMB were reported in EFSA (2012) with DT_{50}s noted in the range of hours to a few days. Based on a manometric respirometer study EMB was noted to be not readily biodegradable (0% degradation, 28 days; EFSA, 2012). Two simulation studies are available in sediment/water systems, but one was deemed not reliable as the sediment content was too low and adherence of EMB to the walls of the test vessel were reported (EC, 2011). The second study reported a DT_{50} in water for dissipation of 8.7 days in both a sandy loam system and sand system. This value is noted to largely reflect the partitioning of the substance to the sediment rather than degradation. The DT_{50} for sediment was reported as >120 days, with the overall DT_{50} for the water/sediment system >120 days (EFSA, 2012). The maximum level of EMB in sediment was noted as 71.3% and 83% after 90 and 120 days (EC, 2011). A further simulation study considered degradation of EMB and deltamethrin (and their respective commercial formulations) over 135 days at 4 and 10 °C in field collected sediments from the vicinity of an active fish farm. This study found that whilst deltamethrin (from its commercial formulation and the substance itself) degraded slowly at 10 °C, EMB and its commercial formulation did not degrade at either temperature under abiotic and biotic conditions (Benskin *et al*, 2014). A recent study looked at biodegradation of active substances present in a field-collected marine "flocculent" sample from the vicinity of an active fish

farm in Canada at 4 °C over 96 days (Hamoutene and Salvo, 2020). The EMB detected in the sample was thought to have been present for around 3 months prior to the time of sampling, based on the last usage of the substance at the farm. The median measured concentration noted in the marine flocculent in this study was higher than a previous study at the site in 2016 (16.5ng/g vs 2.8ng/g). The study found degradation of organic matter in the sample was not accompanied by chemical degradation, with an estimated half-life for EMB of 404 days. The authors concluded that their study showed that laboratory tests that do not take into account weathering may underestimate environmental half-lives for some substances, but note also that weathering may decrease bioavailability and that further laboratory trials that mimic weathering within sediments are needed to better understand degradation pathways.

2.5 Bioaccumulation

Although the octanol-water partition coefficient of log Kow 5 at neutral pH indicates the potential for aquatic bioaccumulation, EMB has a low measured steady state whole fish BCF of 82 L/kg in bluegill fish (*Lepomis macrohirus*; EC 2011 and EFSA 2012).

3 Effects Data

3.1 Human toxicological data

Based on the NOAEL of 0.25mg/kg noted for a 104-week study in rats, and 14-week and 52-week studies in dogs, an ADI of 0.0007mg/kg bw/day was proposed (expressed as EMB) (EFSA, 2012).

The EU harmonised CLP classification for EMB indicates that it has not been classified as carcinogenic, mutagenic or toxic for reproduction (EU, 2021).

3.2 Ecotoxicological Data

3.2.1 Laboratory ecotoxicity data

EMB has a well investigated mode of action, involving binding to *gamma* aminobutyric acid receptors (GABA receptors) and glutamate gate chloride channels with subsequent disruption of nerve signals in arthropods, so particularly relevant for crustaceans, insects, nematodes and tardigrades, but also molluscs and platyhelminths ((Wolstenholme 2012, Lynagh *et al* 2015).

Acute and chronic aquatic toxicity data are available for EMB for a range of freshwater and saltwater species including algae, invertebrates and fish. The data collated for EMB are summarised in Annex 1.

Based on the approach described in section 1.3 the key reliable and relevant acute and chronic ecotoxicity data for freshwater and marine pelagic organisms (ie those assigned a reliability score of 1 or 2) are summarised in Tables 1 and 2 respectively. These studies are highlighted in bold within Annex 1.

Table 1: Reliable acute and chronic ecotoxicity data for freshwater pelagic organisms

Species	**Test duration**	**Endpoint**	**Result (µg/L)**	**Comment**	**Reference**
Acute toxicity data					
Primary producers					
Pseudokirchneriella subcapitata	96h	EC_{50} (growth)	9.65	Mean of 2 studies following principles of OECD 201 (96hr EC50 values of 12.1 µg/l and 7.2 µg/l.	Cited in SEPA (2017) : - EFSA (2009) cited Maynard (2003a) and

					EFSA (2012), EC (2011)
Lemna gibba	14 d	EC_{50} (abundance)	>94	Study used in EFSA (2012)	Cited in SEPA (2017): - EFSA (2012); US EPA (2009); ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)
Crustaceans					
Daphnia magna	48h	EC_{50} (immobilisation)	1.63	Mean of 4 studies following principles of OECD 202 (48h EC_{50}s of 1 µg/l, 1 µg/l, 1 µg/l and 3.5 µg/l).	Cited in SEPA (2017):- WRc (2000); EFSA (2009) cited Blankinship *et al* (2002). EC (2011)
Insects					
Aedes albopictus	24h	LC_{50}	90	Non-standard test species	Cited in SEPA (2017):- Khan *et al* (2011)
Fish					
Oncorhynchus mykiss	96h	LC_{50}	177	Mean of 2 studies following principles of OECD 203 (LC50 values of 174 µg/l and 180 µg/l).	Cited in SEPA (2017):- WRc (2000); EFSA (2012); EC (2011), US EPA (2009)
Pimephales promelas	96h	LC_{50}	194		Cited in SEPA (2017):- Environment Canada (2005), EFSA (2009), EC (2011)
Lepomis macrochirus	96h	LC_{50}	180		Cited in SEPA (2017):- Environment Canada (2005) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992), EFSA (2008)
Chronic					
Primary producers					
Lemna gibba	14 d	NOEC (abundance)	94	Study reported in EFSA (2012)	Cited in SEPA (2017):- EFSA (2012); US EPA (2009); ECOTOX (2016)

					citing US Pesticide Ecotoxicity Database (1992)
Crustaceans					
Daphnia magna	21 d	NOEC (reproduction)	0.088	Study following principles of OECD 211	Cited in SEPA (2017):- Environment Canada (2005) EC (2011)
Fish					
Pimephales promelas	32 d	NOEC (hatching success, survival and growth)	12		Cited in SEPA (2017):- WRc (2000)
Pimephales promelas	32 d	NOEC (reproduction; growth)	6.5		Cited in SEPA (2017):- US EPA (2009) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)

The most sensitive species in both acute and chronic freshwater studies is the invertebrate *Daphnia magna*, with a 48-hour EC_{50} of 1.63 µg/L (mean, four studies) and a 21-day NOEC for reproduction of 0.088 µg/L.

Table 2: Reliable acute and chronic ecotoxicity data for marine pelagic organisms

Species	**Test duration**	**Endpoint**	**Result (µg/L)**	**Comment**	**Reference**
Acute					
Crustaceans					
Crangon crangon	192 h	LC_{50}	166		Cited in SEPA (2017):- WRc (2000)
Acartia clausi	48 h	EC_{50} (immobilisation)	0.28	Copepodite lifestage	Cited in SEPA (2017):- Willis and Ling (2003)
Pseudocalanus elongatus	48 h	EC_{50} (immobilisation)	0.12	Nauplii lifestage	Cited in SEPA (2017):- Willis and Ling (2003)
Temora longicornis	48 h	EC_{50} (immobilisation)	0.23	Nauplii lifestage	Cited in SEPA (2017):- Willis and Ling (2003)
Oithona similis	48 h	EC_{50} (immobilisation)	15.86	Copepodite lifestage	Cited in SEPA (2017):- Willis and Ling (2003)
Americamysis bahia (Mysidopsis bahia)	96 h	LC_{50}	0.059	Mean of 2 studies. (96hr LC50 values of 0.078 µg/l and 0.04 µg/l).	EC (2011); EPP (2018) ECHA (2018)

Nephrops norvegicus	192 h	LC_{50}	572		Cited in SEPA (2017):- WRc (2000)
Molluscs					
Crassostrea virginica	96 h	EC_{50} (immobilisation)	490		Cited in SEPA (2017):- Environment Canada (2005) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)
Crassostrea virginica	96h	NOEC (shell deposition)	260	Embryo test; but considered not a truly chronic study based on exposure duration.	Cited in SEPA (2017):- WRc (2000)
Fish					
Cyprinodon variegatus	96 h	LC_{50}	1430		Cited in SEPA (2017):- WRc (2000) also cited in Environment Canada (2005), EFSA (2009) cited 1995 data, EC (2011)
Chronic					
Crustaceans					
Acartia clausi	8 d	NOEC (fecundity)	0.05	Adult life stage	Cited in SEPA (2017):- Willis and Ling (2003)
Americamysis bahia	28d	EC_{10} (reproduction)	0.00944		EPP (2018b)
Americamysis bahia	28d	NOEC (growth)	0.0087		US EPA (2009), ECOTOX (2016)

The most sensitive species in both acute and chronic marine studies is *Americamysis bahia*, the mysid shrimp with a 96-hour LC_{50} of 0.059 µg/L (mean of two studies) and 28-day EC_{10} (growth) of 0.0087 µg/L. A more recent study reports an EC_{10} for reproduction with a similar effect level (0.0094 µg/L). In this more recent study, several endpoints were measured; two of the other endpoints gave slightly lower results, which were lower than the key study in this species. The statistical significance for use in hazard assessment however of one of these results was unclear (NOEC of 0.00413 µg/L for female body weight, reported at a significance level 1% rather than 5%) and the other was for a secondary endpoint (NOEC of 0.00784 µg/L for mortality in the G2 generation at day 28), so the result for reproduction was taken forward as the critical endpoint for hazard assessment from this study.

Sediment toxicity data for EMB were located for a range of organisms including both freshwater and saltwater species. Acute and chronic data were available for saltwater but only chronic data were located for the freshwater environment. Tables 3 and 4 summarise the available reliable (ie assigned reliability scores of 1 or 2) and relevant ecotoxicity data for sediment dwelling organisms in freshwater and marine water respectively.

The sediment toxicity studies vary in terms of the organic carbon (OC) content of the sediment. This can influence the result of the toxicity test and therefore the EQS guidance proposes that normalisation of the toxicity data to a standardised sediment with an OC of 5% is undertaken when deriving an EQS for sediment (EU, 2018).

Table 3: Reliable ecotoxicity data for freshwater sediment-dwelling organisms

Species	**Test duration**	**Endpoint**	**Result (µg/kg)**	**Result (µg/kg) dwt normalised to 5% OC**	**Comment**	**Reference**
Acute toxicity data						
No available studies						
Chronic toxicity data						
Insects						
Chironomus riparius	28 d	NOEC (emergence)	1.25 (dwt)	2.72	Sediment OC content 2.3%	Cited in SEPA (2017):-EC (2011); EFSA (2012)
Chironomus dilutus	62 d	NOEC (female emergence rate)	2.7 (dwt)	4.82	Sediment OC content 2.8%	Bradley (2005a)
Crustaceans						
Hyalella azteca	42 d	NOEC (survival, growth and reproduction)	32 (dwt)	43.2	Sediment OC content 3.7%. No effects at highest test concentration	Bradley (2005b)

No acute studies for freshwater sediment dwelling organisms are available. The most sensitive species, and endpoint, was the midge *Chironomus riparius* with a 28d NOEC for emergence of 2.72 µg/kg on a dry weight basis normalised to 5% organic carbon. A study in a different midge species, *Chironomus dilutus*, reported a similar NOEC for a related endpoint. No effects were noted in a study with the crustacean *Hyalella azteca* at the concentrations tested.

Table 4: Reliable ecotoxicity data for marine sediment-dwelling organisms

Species	**Test duration**	**Endpoint**	**Result (µg/kg)**	**Result (µg/kg) dwt normalised to 5% OC**	**Comment**	**Reference**
Acute toxicity data						
Annelids						
Arenicola marina	10 d	LC_{50}	40.8 (dwt)	1020	0.2% OC content sediment	EPP (2018d)
Hediste diversicolor	10 d	LC_{50}	2280 (dwt)	2850	Ca. 4% OC content sediment	Mayor *et al* 2008
Crustaceans						
Corophium volutator	10 d	LC_{50}	141.5 (dwt)	2211	0.32% OC sediment content	EPP (2018c)

Corophium volutator	10 d	LC_{50}	255 (dwt)	319	Ca. 4% OC content sediment	Mayor *et al* 2008
Pandalus platyceros	8 d	EC_{20} (mortality,)	400 (dwt)		Sediment OC <0.5%	Veldhoen *et al* (2012)
Homarus americanus	10 d	LC_{50}	330 (dwt)		Limited details of sediment characterisation (field collected)	Daoud *et al* (2018)
Chronic toxicity data						
Annelids						
Hediste diversicolor	28 d	NOEC (survival, growth)	283 (dwt)	615.2	No effects at highest test concentration. Sediment OC content 2.3%.	Fox (2019)
Nereis virens	30 d	NOEC (growth rate)	240 (dwt)	n/a –sand only exposure	Only 1 test concentration, result indicative only	McBriarty *et al* (2018)
Crustaceans						
Leptocheirus plumulosus	28 d	EC_{10} (growth rate; mean weight per surviving adult)	17.6 (dwt)	275	0.32% OC sediment content	EPP (2018e)
Leptocheirus plumulosus	28 d	EC_{10} (growth rate)	52.8 (dwt)	880	Mean of female and male growth rates (49 and 57 µg/kg dwt, respectively) 0.3% OC sediment content.	EAG (2018)
Leptocheirus plumulosus	28 d	EC_{10} (reproduction)	43 (dwt)	716.7	0.3% OC sediment content	EAG (2018)
Corophium volutator	28 d (&75 d)	NOEC (survival, growth, reproduction)	61.28 (dw)dwt)	53.3	No effects at highest test concentration. Sediment 5.75% OC content.	Scymaris (2018)
Homarus americanus	30 d (study extended to 71d)	NOEC (growth)	45 (dwt)		Interstage growth endpoint. Limited details of sediment characterisation (field collected)	Daoud *et al* (2018)
Homarus americanus	30 d (study extended to 71d)	NOEC (behaviour)	<11.6 (dwt)		Behaviour endpoint (position on back). Limited details of sediment characterisation (field collected)	Daoud *et al* (2018)

Acute and chronic data are available for marine sediment dwelling organisms. In acute studies the lugworm *Arenicola marina* is the most sensitive species with a 10-day LC_{50} of 40.8 µg/kg (dry weight). However, when this result is normalised to an organic carbon (OC) content of 5%, this result increases to 1020 µg/kg (dry weight) as the study sediment had a low organic carbon content of 0.2%. A further acute study for this species is available with a 10-day LC_{50} of 111 µg/kg (wet weight) but no details of sediment OC content are available (see Annex 1). When considering results normalised to a standard sediment organic carbon content of 5%, the crustacean *Corophium volutator* is the most sensitive, with a 10-day LC_{50} of 319 µg/kg (dry weight). This result is for a test that was run with a sediment organic carbon content of about 4% (Mayor *et al*, 2008). A separate, comparable, acute study with the same species was run with a sediment containing only about 0.32% organic carbon (EPP, 2018c). The "face value" (that is, as reported before any normalisation to improve comparability between studies) result from this study was similar to that from the study by Mayor *et al* (141.5 vs 255 µg/kg dry weight), but when the result is normalised to 5% OC the result is 2211 µg/kg (dry weight), nearly seven times higher than the Mayor *et al* (2008) study (see discussion below). Acute studies are also available for the shrimp *Pandalus platyceros* and the lobster *Homarus americanus* with similar "face value" results as for *C. volutator*, however these lack detail on exact OC content of the test sediments so cannot be compared on this basis.

In the chronic dataset two studies with annelids are available, ie the ragworm *Hediste diversicolor* and the clam worm *Nereis virens*. The ragworm study showed no effects at the highest test concentration for both endpoints, ie growth and survival (283 µg/kg dwt; 615 µg/kg dwt normalised to 5% OC). The clam worm (*Nereis virens*) study showed effects on growth rate, however only one test concentration of 240 µg/kg was used in this study so it is not possible to use this to derive a threshold for effect. There are chronic studies available for three crustacean species, *Leptocheirus plumulosus*, *Corophium volutator* and *Homarus americanus*. Of these the most sensitive at "face value" is for *L. plumulosus*, with an EC_{10} of 30.5 µg/kg dwt for growth rate, derived from the mean result from two separate studies. When normalised to 5% OC, this result is 492 µg/kg dwt.

A study on the lobster *H. americanus* gave a NOEC for growth of 45 µg/kg. High levels of mortality were observed in EMB treatments in this study, with the behavioural endpoint "positioned on back" being a precursor to this (death was observed after 1 – 2 days for all animals position on back, PoB). A NOEC for PoB could not be derived from this study, as effects were seen for this endpoint at the lowest test concentration (8.8 µg/kg wwt). Although not reported, it should be possible to derive a 30-day EC_{10} for mortality from this study if the data were available (reading from a graph, this would be around 20 µg/kg wwt). The results in this study show that for sublethal endpoints the American lobster is sensitive to EMB.

Comparing the acute and chronic datasets, there are acute and chronic studies for the ragworm *H. diversicolor* while there is no comparable chronic study for the lugworm *A. marina*. In the acute dataset, *A. marina* appears to be more sensitive than *H. diversicolor*, based on the reported LC_{50} and the OC normalised values. The lugworm is a detritivore whereas the ragworm is an opportunistic feeder, with a potentially more varied diet, eating invertebrates as well as detritus (Gerino *et al*, 2003). These functional differences may account for the difference in observed sensitivities, as stated in Mayor *et al* (2008), although the authors do state this cannot be concluded from their study in isolation.

No effects were seen in the *H. diversicolor* chronic study, which was run with a maximum exposure concentration about 4.5 times lower than the LC_{50} from the acute study. Similarly for *C. volutator*, no effects were seen in the available chronic study with a similar maximum exposure concentration

relative to the acute LC_{50} (based on the more sensitive of the two acute tests that had a similar OC content to the chronic study). The study for *H. americanus* that includes acute and chronic endpoints indicates this species is sensitive to EMB, although it cannot be compared like-with-like with the other studies as results are not normalised for OC content as information on the OC content of the field collected test sediment was not available.

Differences in sediment OC content makes comparisons between studies difficult even when results are normalised to a standard OC content. It is possible OC has an influence on the level of toxicity observed, depending on the species, that normalisation to a standard OC content doesn't account for. The one example here that suggests this may be an issue is for the two acute studies with *Corophium volutator*, where one study had a low OC of 0.32% and the other an OC content of 4% which is near the default value of 5% noted in the EQS Technical Guidance (EU 2018). Whilst both studies had similar nominal LC50 results (141.5 and 255 µg/kg dry weight), normalisation to 5% OC greatly changes this (2211 vs 319 µg/kg dry weight). Cheng *et al* (2020) have suggested that low OC lowers bioavailability; this theory is compatible with the results for these two studies when reported on an OC-normalised basis. Others (see Mayor *et al* 2008 and references therein) state that it is quite possible that higher OC content of test sediments enhances bioavailability of test substances, but that other factors including pH and its effect on sulphide and ammonia concentrations, temperature and salinity may be responsible for differences seen in observed levels of toxicity for this species.

More generally, exposure conditions in marine sediment toxicity tests may differ to those of freshwater sediments due to differences in ionic strength and sediment characteristics (for example, clay mineral composition and hence binding/bonding interactions). It is difficult to gauge how much of an effect this may have on bioavailability. Due to precipitation processes which occur in estuaries at the freshwater/marine water interface clay mineral content of marine sediments and active binding capacity, particularly in estuarine regions, is likely to be higher in marine sediments. This is an additional difference between freshwater and marine sediment studies, besides other differences such as feeding behaviour and whether organisms inhabit sediments or live in burrows (which can also influence exposure to chemicals such as EMB).

3.2.2 Meso- and Microcosm studies

A reliable freshwater microcosm study is available (EC, 2011, Volume 1). This 139-day study involved exposure to emamectin benzoate via three applications spaced at 7-day intervals with no recirculation of system water. The study reported a No Observed Ecologically Adverse Effect Concentration (NOEAEC) of 3 * 0.3 µg/L. A saltwater microcosm study (Cheng *et al* 2020) that included *Arenicola marina*, *Corophium volutator* and *Cerastoderma edule* is also available, but because of high levels of mortality in the controls this study was deemed unreliable for hazard assessment. See Annex 1 for more details on these studies.

3.2.3 Field studies

Three field studies are available, all of which follow generally accepted scientific principles of realworld monitoring and analytical studies. One study was conducted by SEPA at eight fish farms on the Shetland Isles (SEPA 2018), a second "passive" field study conducted by the Scottish Association for Marine Science (SAMS) on behalf of industry at fish farms in the Outer Hebrides, West Coast, Shetland and Orkney (SAMS 2018), and a third "active transport" study, again conducted by SAMS on behalf of industry at three active fish farms in the west of Scotland between July 2016 and February 2019 (SAMS 2019). These two studies are described as "passive" and "active" to reflect their purpose. The former had the objective to compare EMB levels present in sediments from different water bodies with a history of varying SLICE® use; whereas the latter involved a long-term field sampling programme with the primary objective of tracking EMB transport to reference stations at fish farms after treatment

with EMB, to understand associated effects on the benthic crustacean community. These studies are briefly discussed below.

The SEPA study involved sampling two or three transects at eight fish farms with "matched" benthic fauna and chemical sediment residue analysis in early 2017 in the Shetland Isles. Based on the results SEPA produced a statistical analytical report (SEPA 2018) along with a summary of their generalised linear mixed modelling (GLMM) data analysis and Canonical Correspondence Analysis (CCA) that was published in a peer reviewed journal (Bloodworth *et al* 2019). Two or more reference sites were included for each fish farm's sampling stations, and hydrodynamic modelling was used to derive transect length and assign sample sites to this transect. Each fish farm had seven to 14 sample stations. The total number of stations, including reference sites was therefore 86. GLMM analysis showed that EMB concentration had the biggest effect on crustacean abundance and number of crustacean species (other parameters considered included total organic carbon, particle size, position relative to predominant flow direction and enrichment of polychaete abundance (SEPA 2018). The statistical analysis was independently reviewed by Biomathematics and Statistics Scotland (BioSS).

Also in 2017 a field study was undertaken on behalf of industry to look at concentrations of EMB in marine sediments in the vicinity of fish farms in relation to indicators of impacts in benthic fauna ("passive" field study; SAMS 2018). Nineteen fish farms in the Outer Hebrides, West Coast, Shetland and Orkney were surveyed based on consideration of historical SLICE® use. Sampling was conducted between July and October 2017. The parameters measured were similar to the SEPA study, although the study design differed in that most sampling stations were further from cage edges. There were up to 10 stations per farm which included a reference station as well as sites at multiple distances outside the fish farm cage edge, mostly beyond the "allowable zone of effect", AZE. In total there were 180 sampling stations. Overall the level of "noise" in the data was high, likely related to the high number of sites & stations and variability resulting from the differing geographies in the sampled fish farms. This had the effect of making interpretation and isolation of potential effects from variables difficult. Nevertheless, as expected EMB concentrations were lowest furthest from fish farm cage edges and increased the closer the station was to the fish farm. Macroinvertebrate species diversity and abundance varied greatly across the survey, although an impact was demonstrated with the extremes of the data. Notably, species diversity and abundance varied greatly in samples with no EMB detected above the limit of detection, presumably a product of the regional variability in environmental conditions of the various farms and regions surveyed. The authors found an apparent relationship between EMB concentrations and species richness as a decline in richness was observed between the limit of detection (1.5 ng/kg wwt) and around 50ng/kg. Above this concentration no further impact was noted. As this relationship seemed unlikely (generally it should be possible to locate a "point of departure" and at concentrations above this observe steadily increasing impacts), the authors "truncated" the data to exclude concentrations less than 0.01 µg/kg and greater than 1 µg/kg wet weight for further statistical analysis, however this approach showed no relationship between species richness and EMB concentration. The authors did find an apparent relationship between particle size and species richness, and less so for organic carbon and species richness, so it appeared environmental factors such as these will have contributed to the lack of clear conclusions from the study.

In an "active transport" study conducted between October 2016 and February 2019, SAMS studied the movement of EMB in the marine environment at three active Scottish fish farms on the west coast of Scotland to distant sampling stations and the associated effects on the benthic community (SAMS 2019). The study captured part or all of 2 growth cycles per farm, with 2-3 EMB treatments in total at each farm. Following "baseline" sampling in July 2016, sampling every other month was carried out for physical parameters (total organic carbon (TOC), particle size analysis (PSA)) as well as

concentrations of EMB (4 replicates from 377 grab samples across the study resulting in 1508 datapoints) at stations. This was done for each of the three fish farms, at the cage edge, two reference stations and a number of stations along a gradient away from cage edges, as well as negative control stations (7 – 9 stations per fish farm in total). Benthic fauna were sampled at the same sites, but at half the frequency of the other parameters (ie once every four months). Total number of individuals, total number of species and species diversity were recorded for each sample. In total there were 16 sampling events, with eight of these including benthic sampling, including the baseline assessment. Benthic data were also truncated to consider a subset of arthropod (mainly crustacean) species with the same indices calculated. The study authors found that EMB was almost always detected (limit of quantification about 1.5 ng/kg wwt), with highest concentrations near cage edges up- and downstream of prevailing currents, although concentrations were variable with the highest variability found in samples near to cage edge. The authors postulated this was a result of a lack of homogeneity in the samples because of the way faecal matter had been deposited. Of the measured concentrations beyond the 100m zone of allowable effect, six out of 1316 replicates were above the current standard for EMB used by SEPA (2020; 0.763 µg/kg wwt). The concentration range within 100m of the cage edge ranged from 0.135 to 5.94 µg/kg wwt. The authors also found that concentrations distant from farms appeared to be at steady state, postulating that enrichment was not occurring as EMB additions were counteracted by removal/dispersion processes. In terms of the biology, there was an overall scarcity of crustacea which made statistical analysis difficult. Highest species richness tended to be found just beyond the cage edge, with a community composition typical of background environment boosted by the presence of opportunist species. Using linear modelling, the authors found a general trend of decreasing species richness with increasing EMB concentration and TOC level, with indicative thresholds for decline above approx 0.8 µg/kg wwt and 3% TOC. It was noted that the overall low species richness meant other factors must be contributing. It was not possible to separate correlations between species richness decrease and TOC enrichment (proximity to fish farm) from those with EMB exposure. Linear modelling found no relationship between median particle size and full community species richness or crustacean species richness at investigated sites. Within the subset of faunal data (the four most commonly occurring species, covering the three amphipods *Leucothoe lilljeborgi*, *Harpinia antennaria* and *Ampelisca tenuicornis*, and the mud shrimp *Calocaris macandreae*) the authors found evidence that different life and feeding strategies influenced sensitivity to EMB exposure. The data indicated a burrowing shrimp species (*C. macandreae*) tolerated moderate to high TOC sediments (up to ca. 5%) but not when EMB concentrations were greater than around 0.25 µg/kg wwt (based on its absence in samples with EMB concentrations above this). Such apparent sensitivity was not seen with the amphipod *A. tenuicornis* which was found in samples with TOC up to about 5% and EMB concentrations up to around 0.75 µg/kg wwt. The remaining two species, *L. lilljeborgi* and *H. antennaria* were absent from sediments with TOC levels above 3 - 3.5% and EMB concentrations above 0.4 µg/kg wwt. The authors described the differences in living/feeding strategies of these four species as a plausible explanation for these observations, summarised as follows. *A. tenuicornis* is a tube building amphipod able to create and control a preferential microenvironment, with its tube physically isolating the individual from sediment contaminants, and is able to switch between suspension feeding and deposit feeding; whereas *C. macandreae* is an exclusively deposit feeding species with a long reproductive cycle and cannot alter feeding strategies. *H. antennaria* is a burrowing amphipod and has a diet mostly composed of crustaceans; *L. lilljeborgi* is a commensal amphipod typically found associated with other taxa although also known to burrow in soft sediment (as a filter feeder exposure to substances behaving like EMB may be lower compared with *C. macandreae*).

3.2.3.1 Further statistical analysis of field data

In 2020 two further publications (Dixon PM 2020a and Dixon PM 2020b) were produced in relation to the above field studies, both relating to reanalysis of the data from SEPA (2018) and SAMS (2018). Both reports were carried out by a consultant statistics expert, commissioned by the industry. The

author reanalysed the two datasets, using (i) a linear mixed model (LMM) with fixed effects for measured environmental variables and a random site effect to represent all unmeasured site characteristics (with environmental variables chosen by model selection) and (ii) a generalised additive mixed model (GAMM) to allow smooth but non-linear effects of environmental characteristics, along with a random side effect. In both approaches the focal variable, EMB concentration, was added to the models. The author then estimated the strength of the association by the estimated regression slope for EMB concentration, and by estimating the difference between two hypothetical sites differing only in EMB concentration, for both datasets and the datasets combined. This approach ignores the stratification inherent in both studies because of the way sites are selected and their relative proximity to one another for each fish farm. One report (Dixon PM 2020a) describes the general statistical approach and results for the two datasets separately, while the second report (Dixon PM 2020b) looks solely at geographical subsets of the SAMS (2018)/SEPA (2018) study. A summary of this review is included in Annex 3.

As stated, the reanalysis undertaken by Dixon (2020a and 2020b) used the situation where two hypothetical sites derived from the best fitting models of the two modelling approaches, differing only in EMB concentration and species richness, are compared for the effect of a doubling of the former on the latter. For the SAMS (2018) study data Dixon concurred with the findings of the original report in that it found no evidence for a relationship between EMB concentration and crustacean species richness. For this dataset the GAMM and LMM were equivalent (so GAMM was not considered further). Estimated regression slopes were shallow, with the least and most negative slopes indicating that a hypothetical site with twice the EMB concentration relative to baseline had an estimated 2% or 2.8%, respectively, fewer crustacean species. The author stated that the effect of EMB concentration is closer to zero than the effects of other variables (TOC, % gravel, % sand, % mud); they were not statistically significant. On the other hand reanalysis of SEPA (2018) study data showed more negative estimated regression slopes, statistically significant from zero; using the most and least negative slopes for a hypothetical site, a doubling in EMB concentration relative to baseline would result in 10.4% or 9.4%, respectively, fewer crustacean species. In this case EMB concentration has the largest effect on species richness (followed by TOC, % mud, mean particle size). Analysis of geographic subsets of the passive monitoring field study (seven West Coast sites; 6 Western Isle sites; 6 Orkney/Shetland sites; 5 Shetland sites; SAMS 2018) found no evidence for a relationship between EMB concentration and crustacean species richness (Dixon PM, 2020b). Regression coefficients were close to zero or positive for the West Coast and Western Isles, respectively, and were not statistically significant. Analysis of the combined Shetland SAMS 2018/SEPA 2018 datasets led to statistically significant coefficients, with EMB concentrations accounting for the largest negative effect on crustacean species richness along with % sand (see Table 5).

Table 5: Shetland SAMS 2018/SEPA 2018 combined datasets - Estimated effects by variable on the log crustacean species richness scale for each variable in the most appropriate linear mixed model for each dataset (reproduced from Dixon PM 2020b)

variable	Percentiles of X		Predicted log Scrust		change	95% CI
(X)	25th	75th	25th	75th		
% TOC	0.5	1.782	0.034	-0.029	-0.063	(-0.369, 0.24)
% sand	54.78	87.9	-0.633	-1.016	-0.383	(-0.676, -0.1)
% mud	7.07	33.6	-0.057	-0.272	-0.215	(-0.499, 0.071)
EMB	0.036	0.406	0.49	0.133	-0.357	(-0.543, -0.168)

Estimated by predicting the mean log crustacean species richness for two samples, one with that variable at its 75th percentile and the second at its 25th percentile, then calculating the difference. The 95% confidence interval is calculated from the uncertainty in the regression coefficient for each variable.

3.2.3.2 Discussion of field data

All three studies appear to have been well conducted and clearly described. In the case of the SEPA 2018 study, the data and approach resulted in a peer reviewed publication (Bloodworth *et al* 2019), although aspects of this were critiqued by another statistical expert (see above and Annex 3). There are obvious differences in approaches to site selection and sampling, and statistical treatment of the data, most notably between the SEPA 2018 study and the SAMS 2019 active transport study in terms of the last of these; the latter uses univariate statistical analysis as opposed to for example linear mixed effect models. The datasets collected here are very powerful as they cover a significant period of time, and using such approaches could have added a lot to the findings; for example, the 0.8µg/kg wwt threshold seems to have been estimated visually from a graph of all the data but multi parameter linear modelling could have been used to properly assess the effect of EMB in relation to other likely influential variables. Whilst more detailed analysis of the data would most likely not make a material difference in the findings, it could give more confidence in the findings. The authors suggest there is co-linearity between EMB and total organic carbon (TOC), but they do not try to assess the difference in effects between the two. The variable sensitivity of different crustacean species to EMB was similarly found in the SEPA 2018 study. Interpretation of benthic impacts due to chemicals is difficult, which is why sediment quality TRIAD studies in which bioassays are incorporated into the assessment are often recommended although such approaches were not used in the reported studies and would likely have been impractical.

The statistical reanalysis undertaken by Dixon (2020a and 2020b) and described above looked at both SEPA's Shetland data and the data from the SAMS 'passive' field study using similar techniques to Bloodworth *et al* (2019). However, it did not assess the impact of predictor variables on benthic community composition using canonical correspondence analysis as was done in Bloodworth *et al* (2019). Given the noted limited overlap in community composition between the Orkney and Shetland datasets and the remaining areas, this omission of multivariate modelling means it is not understood if crustacean species richness was necessarily the most appropriate response variable for a GLMM. For instance it is known from Wilding *et al* (2017) and Bloodworth *et al* (2019) that lifestyle and functional traits of crustacean species are important in determining how exposed they might be to sediments with high concentrations of EMB. It would be useful to look into this in more detail in order to understand if in a larger dataset, the key relationship between EMB and crustaceans is more pronounced according to feeding method, mobility and other functional traits. Also, the data could be used to explore futher whether the relative absence of some crustacean taxa open a niche for other non-crustacean species that might fulfil a similar functional role (and whether harm to crustacean species reduces overall complexity in the assemblage of species as a whole).

In general, the reanalysis of the SEPA study came to the same conclusions as Bloodworth *et al* (2019), albeit with a lower significance of effect due to differences in data handling. The same correlation between EMB exposure and species richness was not found in the data from the SAMS passive study in the Western Isles and West Coast.

The statistical methods employed by Dixon were comparable to the GLMMs applied in Bloodworth *et al* (2019). However, data pre-treatment differed. Bloodworth *et al* (2019) assessed the input variables for multicollinearity using the Variable Inflation Factor (VIF) test. They took this step as collinear variables have the potential to cause model convergence issues, bias in regression coefficients and inflate standard errors (Harrison *et al*, 2018). The use of VIF was based on recommendations made in

Zuur *et al* (2010; see Annex 3). Without assessing collinearity between predictor variables it is possible the issues identified are present in the models of the SAMS study.

As stated above, more detailed analysis of the data would most likely not make a material difference in the findings, although this could potentially give more confidence in the findings.

Overall, two of the three available field studies presented information that the authors interpreted as suggesting a correlation between EMB concentration or TOC and species richness. This was not the case for the passive monitoring field study (SAMS 2018). The SEPA 2018 study suggested a relationship between increasing EMB concentration and a decline in species richness, although it was not possible to derive a threshold for effects from the data (visual inspection of plotted concentration data suggest that a concentration somewhere in the region 0.01 – 0.1ug/kg dwt should be protective of impacts on macroinvertebrate abundance/diversity of benthic fauna), while the active transport (SAMS 2019) study notes a "drop off' in crustacean diversity at an EMB concentration of about 0.8 µg/kg wwt (ca 1.1 µg/kg dwt, assuming a moisture content of 25%), as well as a relationship between absence of a potentially more vulnerable species and EMB concentrations above about 0.25 µg/kg wwt (ca 0.33 µg/kg dwt, assuming a moisture content of 25%). It is possible that no clear relationships could be divined in the passive monitoring field study because of differences in study design (lower density of sampling points) and the way EMB concentration ranges and species presence happened to occur in the analysed samples (variability in the data and differences in background environments across the regions and many farms), although various statistical approaches were applied to the data to try to account for confounding factors (including a correlation between EMB concentrations and species richness that did not follow a monotonic dose-response relationship and so was discounted by the authors). Confounding factors may include bioavailability of EMB, which may vary with sediment residence time and sediment characteristics amongst other factors, and which laboratory extraction techniques may fail to represent.

4 Derivation of Quality Standards

As discussed in Section 1.2, based on the information on the properties of EMB, QS for surface waters and sediment have been identified for derivation. Biota standards for secondary poisoning and human health were not required to be derived based on the low potential for EMB to bioaccumulate and its level of toxicity to human health, as per the TGD guidance (EU 2018). Due to the fact the only authorised use of EMB in the UK currently is as a veterinary medicine to treat farmed Atlantic salmon, QS have only been considered for the saltwater environment. The following sections describe the derivation of QS for saltwater (MAC and AA) and marine sediment.

There are currently two recommended approaches for QS derivation in surface waters and sediment, the so-called deterministic and probabilistic approaches. In the former, the key datapoint (ie the lowest ecotoxicity result from a reliable and relevant study) in the compartment-specific ecotoxicity dataset is selected and an assessment factor (AF) is applied to it to account for uncertainties that include laboratory to field extrapolation, representiveness (unknown sensitivity of untested taxa), etc. The probabilistic approach can be used where larger datasets are available, where a substance's toxicity profile has been better investigated through laboratory tests representing many taxanomic groups and species. In this approach a distribution of the sensitivities of tested species is plotted relative to common toxicity metrics (for example, NOEC or EC_{10} for chronic toxicity studies) in a Species Sensitivity Distribution (SSD). This is used to derive the concentration that is hazardous for 5% of the tested species (the HC_5). An assessment factor is applied to this HC_5 to derive the QS. The AF is lower than those used in the deterministic approach because levels of uncertainty are lower owing to the more extensive dataset.

4.1 Pooling of Fresh- and saltwater data

The EQS Technical Guidance (EU 2018) notes that if no systematic or statistical differences are apparent between marine and freshwater data, it is appropriate to pool the data for the purposes of QS derivation. Apparent differences in the fresh and marine pelagic datasets for EMB are a consequence of the marked differences in species composition, with a much higher percentage of sensitive taxa in the marine dataset. Statistical analysis of the available pelagic data identified no differences between the fresh- and marine water datasets for pelagic organisms.

There are not enough data to carry out a meaningful statistical comparison of the freshwater and marine sediment ecotoxicity data. In addition there are marked differences in the represented taxa – 2 insect species and 1 crustacean for freshwater chronic studies, versus 2 annelid species and 3 crustaceans for marine chronic studies. The freshwater studies clearly show that insects are far more sensitive to the substance than the one freshwater crustacean (no effects observed in the latter at the concentrations tested). Overall, the sediment toxicity data base does appear to indicate that marine species are less sensitive than freshwater species. This could also reflect a difference in sediment binding characteristics reducing bioavailability, amongst other factors (see section 3.2.1), in addition to physiological differences. In line with EU 2018 however, it is appropriate to pool the available datasets on the basis of no obvious systematic differences between them.

4.1.1 Consultation and Peer Reviewer Responses Relevant to Data Selection & Pooling

In comments submitted to the 2019 consultation several arguments were put forward by some of the respondents as to why freshwater and marine data should not be pooled in the derivation of the QS for sediment, and specifically why insect data should not be used (discussed more fully in section 4.4). Annex 2 includes a fuller summary of the consultation comments.

Three respondents stated that freshwater data should not be included in the derivation of marine EQS for this substance based on a consideration of its toxic mode of action. Specifically, they all stated that ionic gradients across neuronal membranes are very different between fresh water and marine environments and organisms, and so substances like EMB that affect GABA and GluCl receptor ion channel function (Wolstenholme 2012, Lynagh *et al* 2015) could impact freshwater organisms differently to marine organisms. However systematic differences are not apparent when comparing the largest datasets for the substance, the freshwater and marine pelagic acute and chronic datasets. Additionally, in both cases it is a marine species that is more sensitive in acute and chronic tests (mysid shrimp). This suggests that within the available datasets the relative sensitivity of certain groups of organism depends more on their specific physiologies irrespective of whether they are fresh water or marine organisms.

Peer reviewers of this report were specifically asked "*Has the correct approach to data pooling been used (freshwater and marine data have been pooled)?*". The first reviewer agreed with the approach, stating that it is consistent with the recommendations of EQS Technical Guidance (EU 2018) with respect to both pelagic and benthic species. The second reviewer considered the approach acceptable, stating that, while data are available for many different species and so interpretation is difficult, no consistent difference in sensitivity within similar phyla between freshwater and marine species is indicated (for example, although the marine crustacean *Americamysis bahia* appears to be more sensitive than the freshwater crustacean *Daphnia magna,* it appears freshwater fish are more sensitive than marine fish); and that pooling both sets of data provides additional confidence that specific adverse effects are not missed because the combined data set encompasses a greater variety of species and biological endpoints.

4.2 Derivation of MAC-$QS_{sw, eco}$

The available reliable and relevant acute dataset is listed in Tables 1 and 2 for fresh and marine water respectively. It includes 1 freshwater algal species and one plant, 8 crustaceans (1 lobster, 2 shrimp, 4 copepod marine species and 1 freshwater species), 1 marine mollusc, 1 freshwater insect species and 4 fish species (1 marine and 3 freshwater).

There are not enough data to satisfy the requirements for the construction of a species sensitivity distribution (SSD), so the deterministic approach is used to derive the QS. The lowest reliable and relevant result is for the mysid shrimp (*Americamysis bahia*) in two tests conducted according to OPPTS Guideline 850.1035: Mysid Acute Toxicity Test (1996). These studies gave 96h LC_{50}s of 0.078µg/L and 0.04 µg/l, with a mean of 0.059 µg/l.

Based on the EQS Technical Guidance (EU,2018) (See Table 6, section 3.4.2.1), the available acute ecotoxicity dataset for EMB and knowledge of the substance's toxic mode of action, an assessment factor of 50 is proposed for application to the lowest acute effect concentration noted above. This is based on the fact that acute data is available for algae, crustaceans and fish and in addition data is available for one specific saltwater taxonomic group, ie the mollusc *Crassostrea virginica*. Application of an AF of 50 to the acute value of 0.059µg/L results in a MAC-$QS_{sw,eco}$ of 0.00118µg/l, or 1.2ng/l (rounded).

The EQS Technical Guidance (EU, 2018) notes that marine organisms that belong to the taxa algae, crustaceans or fish but have a different life form or feeding strategy than those represented in the freshwater toxicity dataset can be considered additional marine taxonomic groups and may also allow a reduction in the size of the assessment factor applied. A study on the crustacean Norway lobster (*Nephrops norvegicus*) is included in the available dataset for EMB.

In the peer review of SEPA (2017), one reviewer suggested that this species is significantly different from the other crustaceans (copeopods) and has a different feeding strategy. As a result it was proposed that consideration of this data along with the data for *Crassotrea virginica* could justify using an assessment factor of 10 based on the EQS technical guidance. However, one respondent to the consultation in 2019 commented that the choice of this assessment factor may be underprotective for larval stage lobsters. They argued the test may not take into account additional exposure to the substance via sediment ingestion that may occur based on the substance's pattern of use and release (SEPA 2017; WRc 2000). Considering the available dataset (including the *Crassotrea virginica* and *Nephrops norvegicus* acute studies), peer reviewers agreed with the used of an assessment factor of 50, giving a MAC-$QS_{sw, eco}$ of 0.00118 ug/l, or 1.2 ng/l (rounded).

4.3 Derivation of AA-$QS_{sw, eco}$

The available reliable and relevant acute and chronic datasets are listed in Tables 1 and 2. Chronic data include 1 primary producer (freshwater), 3 crustacean species (1 freshwater and 2 marine), and 1 freshwater fish species.

There are not enough data to satisfy the EQS Technical Guidance (EU 2018) requirement for the construction of a species sensitivity distribution, so the deterministic approach will be used to derive the QS. The lowest chronic effects data is from a 28-day mysid shrimp study with a NOEC (growth) of 0.0087µg/L based on US EPA guideline OPPTS 850.1350 (1996). A separate related study reported a similar result but an EC_{10} for reproduction (0.00944 µg/L; this study also reported lower effect results

for other endpoints[1], however issues with these results mean that the EC_{10} for reproduction was the key endpoint).

The dataset includes four reliable chronic studies in freshwater organisms (3 taxa) in addition to the mysid shrimp and *Acartia clausi* marine studies (the oyster study in Table 2 is considered sub-lethal, not a true chronic study). There are acute studies in seven freshwater species and eight marine species including the study on the oyster *Crassostrea virginica*. In acute studies the mysid shrimp is the most sensitive species with a reported 96hr LC_{50} of 0.059µg/L (mean of two studies). The available acute marine dataset indicates that the mysid shrimp is likely to be chronically more sensitive than these other marine species in the acute dataset. The EQS Technical Guidance (EU, 2018, Table 4 section 3.3.2.1, footnote (d)) indicates that an assessment factor of 100 applies where chronic data is available for 3 species across three trophic levels (ie algae, invertebrates and fish), but that this can be lowered to a *minimum* of 10 where acute studies show that additional marine species (eg echinoderms or molluscs) are not the most sensitive group and it has been determined with high probability that in a chronic test these species would be less sensitive than the most sensitive species in the available chronic database. Based on the available acute and chronic datasets for freshwater and marine species, an assessment factor of 50 is most appropriate. This is because chronic data is available for EMB across three species in three trophic levels and in addition an acute study is available for an additional marine species, ie the oyster*, Crassostrea virginica,* which indicates it is not the most sensitive species. Applying an AF of 50 to the lowest chronic NOEC of 0.0087µg/L gives an AA-$QS_{sw, eco}$ of (0.000174µg/l) 0.17 ng/L (rounded).

4.4 Derivation of $QS_{sediment, sw\ eco}$

As discussed in Section 1.2 EMB meets the screening criteria for sediment QS derivation as the log Koc is >3, the log Kow is >3 (See section 2.3) and evidence exists suggesting accumulation in sediments (see section 3.2.3). Previously two marine sediment EQS were derived, one protective of "near field" effects and the other protective of "far field" effects, in line with the approach taken to regulating the substance in Scottish fish farms before more recent changes under SEPA's sector plan (SEPA, 2018a). This approach has been dropped and a single sediment QS has been derived, following the EQS Technical Guidance (EU, 2018) and designed to be protective of the marine environment as a whole.

The relevant and reliable freshwater and marine sediment toxicity data are summarised in Tables 3 and 4. Both acute and chronic toxicity data are available. EQS Technical Guidance (EU, 2018) notes that where sediment toxicity data are available these should be used to derive the QS in preference to the use of the equilibrium approach (to estimate sediment toxicity based on available aquatic toxicity data). Results of long-term toxicity tests with sediment organisms are preferred for deriving sediment standards due to the generally long-term exposure of benthic organisms to sediment bound substances (EU, 2018). As discussed in section 4.1, freshwater and marine data have been pooled. The available reliable and relevant dataset comprises chronic data for two freshwater insect species, two marine worm species, and four crustacean species (1 freshwater and 3 saltwater).

There are not enough toxicity data to construct a species sensitivity distribution for EMB and therefore the deterministic approach is used to derive the QS. The most sensitive study, when the data are

[1]NOEC for female body length of 4.13ng/l, but derived at a significance level of 1%, not 5% as is more usual and no EC10 could be derived for the effect below the highest concentration tested, and there was no effect for the same endpoint for males. As it is unlikely there would be a sex-specific growth difference, the result is uncertain and has not been used here. The next lowest NOEC was 7.84ng/l (EC10 24.52ng/l) for mortality in the G2 generation at day 28. However, this is not stated to be a key endpoint for this test as the EPA do not recommend an MATC is developed for it. The third lowest endpoint, used in this assessment, was an EC10 of 9.44ng/l (95% CI 1.72, 15.01) for reproduction (offspring per surviving female per reproduction day); a NOEC of 17.07 was given for the same endpoint.

normalised to 5% OC, gave a NOEC for emergence of 2.72 µg/kg dry weight for the freshwater midge *Chironomus riparius*. The next most sensitive result was for the freshwater midge *Chirononomus dilutus* with a NOEC of 4.82 µg/kg on the same reporting basis. The most sensitive marine species appears to be the lobster *Homarus americanus*, with a NOEC (for the behavioural endpoint "positioned on back") of <11.6 µg/kg dry weight and a NOEC for growth of 45 µg/kg dry weight, but as the OC content of the field-collected sediment used in this study was not reported the result cannot be compared on the same basis with the other results. The unbounded result for "position on back" adds some uncertainty to the database for sediment, if this is considered to be a population relevant effect.

Based on the available dataset, an assessment factor of 10 applies ("Three long term tests with species representing different living and feeding conditions including a minimum of two tests with marine species"; EC 2018, section 5.2.4.1 Table 13). The criteria are fulfilled in that the available dataset includes four long term tests with species representing different living and feeding conditions (the ragworm *Hediste diversicolor*, the arthropods *Leptocheirus plumulosus* and *Corophium volutator*, the lobster *Homarus americanus* and the two freshwater midge species). This would give a $QS_{sediment,\ sw\ eco}$ of 0.272 µg/kg dwt, or 272 ng/kg dwt when applying the AF of 10 to the lowest NOEC of 2.72 µg/kg dwt for the freshwater midge *Chironomus riparius (normalised to 5% organic carbon)*.

The acute dataset can be used to give context to this derivation from the chronic data. In the acute dataset, the most sensitive species on a dry weight basis is *Corophium volutator* when results are normalised to 5% OC content (LC_{50} = 319 µg/kg dwt). Using an assessment factor of 1000, which would apply in the absence of any chronic data, and the 10-day LC_{50} for *C. volutator*, would give a QS of 0.319 µg/kg dwt. This result is comparable to the $QS_{sediment,\ sw\ eco}$ derived above based on chronic data.

According to the EQS technical guidance (EC 2018) (section 5.2.1.3), field data are important in EQS derivation as lines of evidence to help reduce uncertaintly although they are not usually used as "higher tier" test data (ie in place of laboratory studies). In practice this means field data can be used to consider a QS derived from laboratory data, and in some cases (eg where differences in effect are greater than an order of magnitude) make the case for a different assessment factor. Three field studies are available, with two of these having been reanalysed by a statistical expert (see section 3.2.3). Effects were noted in two of these studies, with the SEPA (2018) study suggesting EMB concentrations in the range 0.01 – 0.1 µg/kg dwt should be protective of impacts on macroinvertebrate abundance/diversity of benthic fauna and the active transport study (SAMS 2019) indicative of impacts on a sensitive species above around 0.25 µg/kg wwt. These results do not provide strong enough evidence to suggest a change from the default assessment factor of 10 as presented above.

Three respondents to the 2019 UKTAG standards report consultation stated that freshwater insect data specifically should not be used in the derivation of a marine QS, because they believe this group of organisms is not relevant for the marine environment. They cited a lack of evidence of presence of insect species in intertidal zones in the vicinity of operating fish farms, whilst two of the respondents state that insect species are rarely or never found in surveys of subtidal benthic fauna in the vicinity of fish farms (carried out for the purposes of regulation). One respondent went on to state that if the purpose of a marine EQS is to protect subtidal benthic faunal communities, then insect data is irrelevant. One respondent of the three also stated that data requirements are met with the marine sediment chronic data alone. Other respondents to the consultation agreed with the derivation. These comments led to questions on what the protection goals of an EQS are, and a key premise of QS setting under WFD around uncertainty in the coverage and representivity of the available dataset for the real world environment. The Water Framework Directive (EC 2000) and the technical guidance for EQS derivation (EC 2018) both indicate that the protection goal for the marine environment is wider than

the subtidal benthic faunal community, being "protective of all types of surface waters and communities". This indicates environments not normally surveyed for the regulation of a fish farm are in scope of a marine EQS (note in this regard an EQS's protection goal doesn't dictate a particular regulatory strategy, nor does it have a bearing on survey strategies for regulation). In Article 1 (Purpose) the WFD states "the purpose of this Directive is to establish a framework for the protection of inland surface waters, transitional waters, coastal waters and groundwater which: (a) prevents further deterioration and protects and enhances the status of aquatic ecosystems and, with regard to their water needs, terrestrial ecosystems and wetlands directly depending on the aquatic ecosystems;…and thereby contributes to: the provision of the sufficient supply of good quality surface water and groundwater as needed for sustainable, balanced and equitable water use; …the protection of territorial and marine waters…". Marine EQS cover transitional and coastal waters. The Directive defines transitional waters as "bodies of surface water in the vicinity of river mouths which are partly saline in character as a result of their proximity to coastal waters but which are substantially influenced by freshwater flows" and coastal waters as "surface water on the landward side of a line, every point of which is at a distance of one nautical mile on the seaward side from the nearest point of the baseline from which the breadth of territorial waters is measured, extending where appropriate up to the outer limit of transitional waters." It is accepted that insects are an under-represented taxonomic group in the marine environment, potentially present only in intertidal zones. Nevertheless, the chronic freshwater insect data are included in the pooled sediment dataset on the basis that a precautionary approach should be taken to setting marine EQS, which are protective of all transitional and coastal waters.

In conclusion, a $QS_{sediment,\ sw\ eco}$ of 0.272 µg/kg dwt, or 272 ng/kg dwt, is recommended.

Although the same test result has been used in this derivation, the QS is about 11.5 times less stringent than the previous derivation that was consulted upon in 2019. This is because the available dataset is much larger than was available for the previous derivation, decreasing the level of uncertainty and so the assessment factor that is applied (from 50 to 10). Normalising the study result to a default sediment organic carbon content that the EQS is derived from accounts for the remaining difference. This follows the rationale and guidance of EU 2018.

4.5 Implications of the Proposed Values for Environmental Quality Assessment

Data on concentrations of EMB in the UK's water environment are generally sparse. The exception is in the regulation of fish farms in Scotland, where sub-tidal sediment data are collected and used in regulation. The standards in Scotland that were derived in 1999 for the regulation of this sector are a surface water MAC of 0.00022 µg/l and a sediment "MAC" of 0.763 µg/kg wet weight (which equates to 954 ng/kg dry weight, assuming a moisture content of 25%). These are listed in a SEPA guidance document as Environmental Quality Standards (table 9a "Operational Water Quality Standards used by SEPA for regulating the use of chemicals in aquaculture"; SEPA 2020); they do not appear in Scottish legislation. As such, the MAC-$QS_{sw,\ eco}$ proposed here, 1.2 ng/l, is about five times less stringent than the 1999 MAC in SEPA's guidance and the $QS_{sediment,\ sw\ eco}$ proposed here of 272 ng/kg dwt is about three and a half times more protective than the 1999 standard (on a dry weight basis). An interim standard of 23.5 ng/kg dwt is also currently being used for new applications (SEPA 2021). The $EQS_{sediment,\ sw\ eco}$ proposed here is about 11.5 times less stringent than this value, for the reasons discussed in section 4.4.

5 Conclusions

Environmental Quality Standards for saltwater and marine sediment have been derived for EMB based on consideration of the available data and the EQS Technical Guidance (EU, 2018). The proposed EQS are summarised in Table 6. Only EQSs for the saltwater environment have been

derived due to the specific use in the UK of EMB as a veterinary medicine for the treatment of sealice in Atlantic salmon.

The derivation has taken into account data from the literature, data provided by the industry as well as comments arising from the consultation on the first UKTAG EQS draft in 2019, and those from two independent peer reviewers of the current report. Ecotoxicity data for a range of freshwater and marine species have been considered, including both acute and chronic data. In addition, data from three field studies undertaken in locations associated with salmon farms in Scotland have also been taken into consideration.

Based on the available data and the EQS Technical Guidance (EU, 2018) both freshwater and saltwater data have been used in the derivation of the EQS as it was considered appropriate to pool the freshwater and saltwater studies. Data for a freshwater species, ie *Chironomus riparius*, drives the derivation of the sediment EQS as it was identified as the most sensitive species from the available dataset. Questions have been raised as to the relevance of an insect species in the marine environment and it is accepted that insects are an under represented taxonomic group in the marine environment, only potentially present in intertidal areas. However, it is considered appropriate to use the insect study on the basis, for example, that an EQS in saltwaters is derived to be protective of all transitional and coastal waters, and there are uncertainties over the data set which lead to the need to a precautionary approach, for example some under represented endpoints with unknown population relevance may be sensitive in saltwater species (study in *Homerus americanus*), and uncertainties over the the effect of variables in saltwater studies including organic carbon content on levels of observed toxicity (studies in *Corophium volutator*). In addition, field studies have indicated potential for effects on organisms starting from concentrations of about 0.1 and 0.25 ug/kg on a wet weight basis. Although there is uncertainty with these values, they give an additional line of evidence that the recommended $EQS_{sediment,\ sw\ eco}$ is set at an appropriate level. Due to the expanded dataset available, it is possible to use the lowest recommended assessment factor of 10 in this precautionary approach.

Peer reviewers accepted the proposed precautionary approach to EQS setting, and the pooling of fresh- and marine water data and the derivation of the EQS for sediment based on chronic freshwater insect data.

The MAC-$QS_{sw,\ eco}$ and the AA-$QS_{sw,\ eco}$ have both been derived based on data for the crustacean *Americamysis bahia* which was the most sensitive species in the available acute and chronic datasets. Assessment factors of 50 have been applied in both cases as insufficient data on additional marine species were available to reduce the assessment factor to 10.

Table 6. Summary of the proposed EQS for emamectin benzoate (EMB)

$EQS_{sediment,\ sw\ eco}$ (ng/kg dwt)	MAC-$EQS_{sw,\ eco}$ (ng/L)	AA-$EQS_{sw,\ eco}$ (ng/L)
272	1.2	0.17
Based on the lowest relevant & reliable study (28-day *Chironomus riparius* NOEC 2720ng/kg dwt, normalised to a sediment organic carbon content of 5%). An assessment factor of 10 was applied; chronic data was available for 2 insect species, 1 polychaete species and 4 crustacean species. Species differences in living/feeding	Based on mean of two lowest relevant & reliable studies (96-hour *Americamysis bahia* LC50s 78 & 40 ng/L). Assessment factor of 50 applied; data available for 2 primary producers, 8 crustacean species, 1 marine mollusc, 2 insect species and 4 fish species. Lack of detail for lobster study and the potential for additional exposure via ingestion justify the use of the	Lowest relevant & reliable study (28d *Americamysis bahia* NOEC 8.7 ng/L). Assessment factor of 50 applied; data available for 1 primary producer, 3 crustacean species and 1 fish species. Differences in living/feeding strategies were considered insufficient to enable use of a lower assessment factor of 10.

strategies supported the use of an assessment factor of 10. Field data provides additional confidence the assessment factor is appropriate and the derived EQS is protective.	assessment factor of 50 rather than a value of 10.	

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Annex 1 summary of ecotoxicity data

Table A1.1: Pelagic Ecotoxicity data

Species	Test duration	Endpoint	Result (µg/l)	Test method details	Comment	Reliability	Reference
Freshwater Acute							
Primary producers							
Pseudokirchneriella subcapitata	5 d	EC_{50} (abundance and growth)	>3.9	Static test. Measured concentrations	Test duration may be too long to ensure exponential growth maintained throughout the study. Original reference not available to check	4	Cited in SEPA (2017) - EFSA (2012), US EPA (2009); ECOTOX (2016) cited US Pesticide Ecotoxicity Database (1992)
Pseudokirchneriella subcapitata	**96h**	**EC_{50} (growth)**	**12.1**	**Static test. OECD 201. Measured concentrations**		**1**	**EFSA (2009) cited Maynard (2003a)**
Pseudokirchneriella subcapitata	**96h**	**EC_{50} (growth)**	**7.2**	**Static test. Mean measured concentration**		**2**	**EFSA (2012), EC (2011)**
Pseudokirchneriella subcapitata	96h	EC_{50} (growth inhibition)	8170	Static test. OECD 201 to GLP, nominal concentration		3	EFSA (2009) cited Wallace (2001a)
Lemna gibba	**14 d**	**EC_{50} (abundance)**	**>94**	**Static test, mean measured concentration**		**2**	**EFSA (2012); US EPA (2009); ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992).**
Invertebrates							
Daphnia magna	**48h**	**EC_{50} (immobilisation)**	**1**	**Flow through study, mean measured concentration**		**2**	**WRc (2000)**
Daphnia magna	48h	NOEC (mortality and immobilisation)	0.3	Flow through study, mean measured concentration		2	WRc (2000)
Daphnia magna	**48h**	**EC_{50} (immobilisation)**	**3.5**	**Flow through study, OECD 202. mean measured concentration**		**1**	**EFSA (2009) cited Blankinship *et al* (2002). EC (2011)**

Daphnia magna	**48h**	**EC_{50} (immobilisation)**	**1**	**Flow through study, mean measured concentration**		**1**	**EFSA (2012); EC (2011), US EPA (2009) refers to 1993 data**
Daphnia magna	**48h**	**EC_{50} (immobilisation)**	**1 (0.84 – 1.2)**	**Flow through study, analysis not reported**		**2**	**Environment Canada (2005) cited ECOTOX (2016) who cited US Pesticide Ecotoxicity Database (1992)**
Daphnia magna	48h	EC_{50} (immobilisation)	>728	Static study, analysis not reported	Result significantly different from 5 other studies. Original study not available for review	4	Environment Canada (2005) cited ECOTOX (2016) who cited US Pesticide Ecotoxicity Database (1992)
Insects							
Aedes albopictus	**24 h**	**LC_{50}**	**90 (40 – 140)**	**Static test, nominal concentrations**		**2**	**Khan *et al* (2011)**
Aedes albopictus	24 h	LC_{50}	1390 - 2450	Static test. Lahore (Pakistan) field population, nominal concentrations		2	Khan *et al* (2011)
Aedes albopictus	24 h	LC_{50}	1350 - 2000	Static test. Faisalabad (Pakistan) field population, nominal concentrations		2	Khan *et al* (2011)
Aedes albopictus	24 h	LC_{50}	1140 - 1700	Static test. Sargodha (Pakistan) field population, nominal concentrations		2	Khan *et al* (2011)
Fish							
Oncorhynchus mykiss	**96 h**	**LC_{50}**	**180**	**Flow through conditions mean measured concentration**		**2**	**WRc (2000)**

Oncorhynchus mykiss	96 h	NOEC (mortality)	87	Flow through conditions mean measured concentration		2	WRc (2000)
Oncorhynchus mykiss	**96 h**	**LC_{50}**	**174**	**Flow through conditions mean measured concentration**		**1**	**EFSA (2012); EC (2011), US EPA (2009)**
Oncorhynchus mykiss	96 h	NOEC (mortality)	49	Flow through analysis mean measured concentration		2	Environment Canada (2005) cited ECOTOX (2016) who cited US Pesticide Ecotoxicity Database (1992)
Pimephales promelas	96 h	NOEC (mortality)	89	Flow through conditions mean measured concentration		2	WRc (2000)
Pimephales promelas	**96 h**	**LC_{50}**	**194**	**Flow through conditions measured concentrations**		**1**	**Environment Canada (2005), EFSA (2009), EC (2011)**
Pimephales promelas	96 h	NOEC (mortality)	160	Flow through conditions analysis not reported		2	Environment Canada (2005)
Cyprinus carpio	96 h	LC_{50}	200	Static test. OECD 203, measured concentrations		3	EFSA (2009) cited Maynard (2003b)
Cyprinus carpio	96 h	LC_{50}	567	Static test. OECD 203, measured concentrations	Test substance commercial formulation A10324A containing 4.89% emamectin benzoate; basis of result and therefore relevance unclear	4	EFSA (2009) cited Wallace (2001b)
Lepomis macrochirus	**96 h**	**LC_{50}**	**180 (40 – 240)**	**Flow through conditions, nominal concentrations**		**1**	**Environment Canada (2005) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992), EFSA (2008)**
Lepomis macrochirus	96 h	NOEC (mortality)	90	Flow through conditions, analysis not reported		2	Environment Canada (2005) cited ECOTOX (2016) who

							cited US Pesticide Ecotoxicity Database (1992)
Freshwater chronic							
Primary Producers							
Pseudokirchneriella subcapitata	5 d	NOEC (abundance)	<3.9	Static test. Analysis not reported	Test duration may be too long to ensure exponential growth phase maintained. Original study not available to check.	4	US EPA (2009) ECOTOX (2016) cited US Pesticide Ecotoxicity Database 1992
Lemna gibba	**14 d**	**NOEC (abundance)**	**94**	**Static test, analysis not reported**		**2**	**EFSA (2012); US EPA (2009); ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)**
Crustaceans							
Daphnia magna	21 d	NOEC (mortality)	88	Flow through study, mean measured concentration	This appears to be the same study as reported below but with units incorrectly reported.	4	WRc (2000)
Daphnia magna	21 d	LOEC (mortality)	160	Flow through study, mean measured concentration	This appears to be the same study as reported below but with units incorrectly reported.	4	WRc (2000)
Daphnia magna	**21 d**	**NOEC (reproduction)**	**0.088**	**Static test, mean measured concentration**	**Likely same study as above**	**1**	**Environment Canada (2005) EC (2011)**
Daphnia magna	21 d	LOEC (reproduction)	0.16	Static test, analysis not reported	Likely same study as above	2	Environment Canada (2005) cited ECOTOX (2016) who cited US Pesticide Ecotoxicity Database (1992)
Daphnia magna	21 d	NOEC (reproduction)	0.088	Static test, mean measured concentration	Possibly same study as above	1	Environment Canada (2005) EFSA, 2009
Daphnia magna	"chronic study" –	NOEC (effect not reported)	0.088	Flow through, analysis not reported	Possibly same study as above	2	US EPA (2009)

	no further details						
Fish							
Pimephales promelas	**32 d**	**NOEC (hatching success, survival and growth)**	**12**	**4 day embryo hatch period and 28 day post hatch juvenile growth period. Mean measured concentrations.**		**2**	**WRc (2000)**
Pimephales promelas	32 d	LOEC (hatching success, survival and growth)	28	4 day embryo hatch period and 28 day post hatch juvenile growth period. Mean measured concentrations.		2	WRc (2000)
Pimephales promelas	32 d	MATC (hatching success, survival and growth)	18	4 day embryo hatch period and 28 day post hatch juvenile growth period. Mean measured concentrations.		2	WRc (2000)
Pimephales promelas	32 d	NOEC/LOEC (growth)	12	Flow through study. Mean measured concentrations.		1	EFSA (2012), EC (2011), ECOTOX (2016)
Pimephales promelas	**32 d**	**NOEC (reproduction; growth)**	**6.5**	**Analysis not reported**		**2**	**US EPA (2009) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)**
Marine acute							
Bacteria							
Vibrio fischeri	5, 15, 30 mins	EC_{50} (bioluminescence)	>6300	Static exposure with measured concentrations	No effect up to max water solubility.	2	Hernando *et al* (2007)
Crustaceans							
Crangon crangon	**192h**	**LC_{50}**	**166**	**Flow through conditions with measured concentrations**		**2**	**WRc (2000)**

Crangon crangon	192h	NOEC (mortality)	<161	Flow through conditions with measured concentrations		2	WRc (2000)
Acartia clausi	48h	EC_{50} (immobilisation)	0.57 (0.04 – 3.99)	Static study. Nauplii life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Acartia clausi	**48h**	**EC_{50} (immobilisation)**	**0.28 (0.1 – 0.69)**	**Static study. Copepodite life stage. Toxicant analysis not reported**		**2**	**Willis and Ling (2003)**
Acartia clausi	48h	EC_{50} (immobilisation)	0.29 (0.08 – 1.1)	Static study. Adult life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Pseudocalanus elongatus	**48h**	**EC_{50} (immobilisation)**	**0.12 (0.07 – 0.2)**	**Static study. Nauplii life stage. Toxicant analysis not reported**		**2**	**Willis and Ling (2003)**
Pseudocalanus elongatus	48h	EC_{50} (immobilisation)	0.14 (0.05 – 0.44)	Static study. Copepodite life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Pseudocalanus elongatus	48h	EC_{50} (immobilisation)	0.45 (0.22 – 0.9)	Static study. Adult life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Temora longicornis	**48 h**	**EC_{50} (immobilisation)**	**0.23 (0.12 – 0.46)**	**Static study. Nauplii life stage. Toxicant analysis not reported**		**2**	**Willis and Ling (2003)**
Temora longicornis	48 h	EC_{50} (immobilisation)	0.41 (0.25 – 0.67)	Static study. Copepodite life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Temora longicornis	48 h	EC_{50} (immobilisation)	2.81 (1.89 – 4.18)	Static study. Adult life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Oithona similis	48 h	EC_{50} (immobilisation)	>15.8	Static study. Nauplii life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Oithona similis	**48 h**	**EC_{50} (immobilisation)**	**15.86 (7.36 – 34.19)**	**Static study. Copepodite life stage. Toxicant analysis not reported**		**2**	**Willis and Ling (2003)**

Oithona similis	48 h	EC_{50} (immobilisation)	232 (64.5 – 13586)	Static study. Adult life stage. Toxicant analysis not reported		2	Willis and Ling (2003)
Americamysis bahia	**96h**	**LC_{50}**	**0.04**	**Flow through conditions, mean measured concentrations. Compound stable throughout.**		**2**	**ECHA (2018)**
Americamysis bahia	96h	NOEC (mortality)	0.018	Flow through conditions, mean measured concentrations. Compound stable throughout.		2	WRc (2000)
Americamysis bahia	96h	MATC (mortality)	0.02	Flow through conditions, mean measured concentrations. Compound stable throughout.		2	WRC (2000)
Mysidopsis bahia (Americamysis bahia)	**96 h**	**LC_{50}**	**0.078 (0.051 – 0.18)**	**Static study. Test guideline US EPA OPPTS 850.1035. measured concentrations**		**2**	**EPP (2018)**
Mysidopsis bahia (Americamysis bahia)	96 h	NOEC (mortality)	0.022	Static study. Test guideline US EPA OPPTS 850.1035. measured concentrations		2	EPP (2018)
Nephrops norvegicus	**192h**	**LC_{50}**	**572**	**Flow through conditions, mean measured concentrations**		**2**	**WRc (2000)**
Nephrops norvegicus	192h	NOEC (mortality)	440			2	WRc (2000)
Lepeophtheirus salmonis	24h	EC_{50} (immobilisation)	243 (127 – 409)	Static study. Salmon and rainbow trout infected with parasites. Parasites collected from a site in		4	Helgesen and Horsberg (2013); ECOTOX (2016)

				an area previously treated with EMB with reported treatment failures. Nominal concentration.			
Lepeophtheirus salmonis	24h	EC_{50} (immobilisation)	302	Static study. Salmon and rainbow trout infected with parasites. Parasites collected from a site in an area previously treated with EMB with reported treatment failures. Nominal concentration.		4	Helgesen and Horsberg (2013); ECOTOX (2016)
Lepeophtheirus salmonis	24h	EC_{50} (immobilisation)	167 (138 – 199)	Static study. Salmon and rainbow trout infected with parasites. Parasites collected from a site in an area previously treated with EMB with reported treatment failures. Nominal concentration.		4	Helgesen and Horsberg (2013); ECOTOX (2016)
Lepeophtheirus salmonis	24h	EC_{50} (immobilisation)	21.5 (18.2 – 23.7)	Static study. Salmon and rainbow trout infected with parasites. Parasites collected from a site in an area previously treated with EMB with reported treatment failures. Nominal concentration.		4	Helgesen and Horsberg (2013); ECOTOX (2016)
Homarus americanus	7 days	LC_{50}	644 µg/g food	Feeding study in adults. Concentrations not reported	Not relevant for watercolumn EQS development.	2	Burridge *et al* (2004)

Homarus americanus	7 days	LC_{50}	>589 µg/g food	Feeding study in stage V and VI juveniles. Concentrations not reported	Not relevant for watercolumn EQS development.	2	Burridge *et al* (2004)
Homarus americanus	≤100 days	EC_{44} (premature moulting)	1 µg/g food	Feeding study in adult females. Concentrations not reported	Not relevant for watercolumn EQS development.	2	Waddy *et al* (2002)
Molluscs							
Crassostrea virginica	**96h**	**EC_{50} (immobilisation)**	**490 (410 – 590)**	**Flow through conditions, concentrations not reported**		**2**	**Environment Canada (2005) cited ECOTOX (2016) citing US Pesticide Ecotoxicity Database (1992)**
Crassostrea virginica	96h	LC_{50}	670	Concentrations not reported		2	Environment Canada (2005)
Crassostrea virginica	96h	NOEC (mortality)	260	Concentrations not reported		2	Environment Canada (2005) cited ECOTOX (2016) who cited US Pesticide Ecotoxicity Database (1992)
Crassostrea virginica	96h	EC_{50} (shell deposition)	530	Flow through conditions. Mean measured concentrations, compound stable throughout	Embryo study of short duration - sublethal	2	WRc (2000) also cited in EFSA (2009)
Crassostrea virginica	96h	NOEC (shell deposition)	260	Flow through conditions. Mean measured concentrations, compound stable throughout	Embryo test; but considered not a truly chronic study based on exposure duration.	2	WRc (2000)
Crassostrea virginica	96h	EC_{50} (shell deposition or embryo larvae)	490	Flow through conditions. measured concentrations not reported	Embryo test; but considered not a truly chronic study based on exposure duration.	2	USEPA (2009)
Fish							
Cyprinodon variegatus	**96h**	**LC_{50}**	**1430 (1250 – 1670)**	**Flow through conditions, mean measured**		**2**	**WRc (2000) also cited in Environment Canada**

				concentrations. Discoloration observed at 500 µg/l			**(2005), EFSA (2009) cited 1995 data, EC (2011)**
Cyprinodon variegatus	96h	NOEC mortality	860	Flow through conditions, mean measured concentrations. Discoloration observed at 500 µg/l		2	WRc (2000) also cited in Environment Canada (2005), EFSA (2009) cited 1995 data, EC (2011)
Marine chronic							
Crustaceans							
Acartia clausi	**8 d**	**NOEC (fecundity)**	**0.05**	**Semi-static study. Adult life stage. Concentrations measured but based on nominal as losses minimal**		**2**	**Willis and Ling (2003)**
Americamysis bahia	28d	NOEC (effect not reported)	0.018	Flow through study. Reported supplemental data, concentrations not reported.		2	US EPA (2009), ECOTOX (2016)
Americamysis bahia	**28d**	**NOEC (growth)**	**0.0087**	**Flow through study. Reported supplemental data, concentrations not reported.**		**2**	**US EPA (2009), ECOTOX (2016)**
Americamysis bahia	28d	LOEC (growth, survival and reproduction)	0.02	Flow through study. concentrations not reported.		2	ECOTOX (2016)
Americamysis bahia	28 d	NOEC (female body weight)	0.00413 (at 1% significance)	Flow through test. Test guideline OPPTS 850.1350. measured concentrations		2	EPP (2018b)
Americamysis bahia	28 d	EC10 (female body weight)	>0.0371	Flow through test. Test guideline OPPTS 850.1350. measured concentrations	Significance at 1% level	2	EPP (2018b)

Americamysis bahia	28 d	NOEC & EC10 (male body weight)	>0.0371	Flow through test. Test guideline OPPTS 850.1350. measured concentrations		2	EPP (2018b)
Americamysis bahia	28 d	NOEC & EC10 (male body weight)	>0.0371	Flow through test. Test guideline OPPTS 850.1350. measured concentrations		2	EPP (2018b)
Americamysis bahia	28 d	NOEC (G2 generation mortality)	0.00784	Flow through test. Test guideline OPPTS 850.1350. measured concentrations	supplemental endpoint for this guideline	2	EPP (2018b)
Americamysis bahia	**28 d**	**EC_{10} (reproduction)**	**0.00944**	**Flow through test. Test guideline OPPTS 850.1350. measured concentrations**	**Value used for hazard assessment from the study**	**2**	**EPP (2018b)**
Echinoderms							
Sphaerechinus granularis	48h	LOEC (development)	1	Static	Few details available for review. short duration test to sensitive life stage developmental effects	4	Sanhueza-Guevara *et al* 2018
Paracentrotus lividus	48h	LOEC (development)	1	Static	Few details available for review. short duration test to sensitive life stage developmental effects	4	Sanhueza-Guevara *et al* 2018
Molluscs							
Choromytilus chorus	48h	LOEC (development)	>1000	static	few details available for review. short duration test to sensitive life stage developmental effects	4	Sanhueza-Guevara *et al* 2018

Table A1.2: Benthic Ecotoxicity data

Species	**Test duration**	**Endpoint**	**Result (µg/kg)**	**Test method details**	**Comment**	**Reliability**	**Reference**

Freshwater chronic							
Insects							
Chironomus riparius	**28 d**	**NOEC (emergence)**	**1.25 (dwt; 2.72 normalised to 5% OC)**	**Static test. OECD 218. Concentrations measured; 94 – 116% of nominal. 1.25µg/kg equivalent to 1.175 to 1.45 µg/kg (measured). 2.3 % OC content**		**2**	**EC (2011); EFSA (2012)**
Chironomus riparius	28 d	NOEC (development)	10 (dwt; 21.74 normalised to 5% OC)	Static test. OECD 218. Concentrations measured; 94 – 116% of nominal. 10µg/kg equivalent to 9.4 to 11.6 µg/kg (measured). 2.3 % OC content		2	EC (2011); EFSA (2012)
Chironomus dilutus	62 d	NOEC (survival)	42 (dwt; 75 normalised to 5% OC)	EPA Test Method 100.5 (2000) and OCSPP Draft Guideline 850.1760 (2009). 2.8% OC. Mean measured concentrations	No effects at highest test conc	1	Bradley 2005a
Chironomus dilutus	62 d	NOEC (growth & emergence)	20 (dwt; 35.7 normalised to 5% OC)	EPA Test Method 100.5 (2000) and OCSPP Draft Guideline 850.1760 (2009). 2.8% OC. Mean measured concentrations		1	Bradley 2005a
Chironomus dilutus	62 d	NOEC (male emergence rate)	5.1 (dwt; 9.11 normalised to 5% OC)	EPA Test Method 100.5 (2000) and OCSPP Draft Guideline 850.1760 (2009). 2.8% OC. Mean measured concentrations		1	Bradley 2005a
Chironomus dilutus	**62 d**	**NOEC (female emergence rate)**	**2.7 (dwt; 4.82 normalised to 5% OC)**	**EPA Test Method 100.5 (2000) and OCSPP Draft Guideline 850.1760 (2009). 2.8% OC. Mean measured concentrations**		**1**	**Bradley 2005a**

Hyalella azteca	**42 d**	**NOEC (survival, growth and reproduction)**	**32 (dw(dwt; 43.2 normalised to 5% OC)**	**EPA test method 100.4 (2000). Mean measured concentrations. 3.7 % OC content**	**No effects at highest test conc**		**Bradley 2005b**
Marine Acute							
Annelids							
Arenicola marina	10 d	LC_{50}	111 (wwt)	Mean measured concentration	no details of sediment characteristics	4	WRc (2000)
Arenicola marina	10 d	NOEC (mortality)	56	Mean measured concentration	no details of sediment characteristics	4	WRc (2000)
Arenicola marina	10 d	MATC (mortality)	76.3	Mean measured concentration	no details of sediment characteristics	4	WRc (2000)
Arenicola marina	**10 d**	**LC_{50}**	**40.8 (dwt; 1020 normalised to 5% OC)**	**Test guideline: ICES Techniques in Marine Environmental Sciences Guideline No. 29. Mean measured concentration. 0.2 % OC content**		**2**	**EPP (2018d)**
Arenicola marina	10 d	NOEC (mortality)	19.9 (dwt; 497.5 normalised to 5% OC)	Test guideline: ICES Techniques in Marine Environmental Sciences Guideline No. 29. Mean measured concentration. 0.2 % OC content		2	EPP (2018d)
Arenicola marina	10 d	EC_{10} (casting)	12.9 (dwt; 322.5 normalised to 5% OC)	Test guideline: ICES Techniques in Marine Environmental Sciences Guideline No. 29. Mean measured concentration. 0.2 % OC content		2	EPP (2018d)
Hediste diversicolor	**10 d**	**LC_{50}**	**2280 (dwt; 2850 normalised to 5% OC)**	**nominal concentrations,no analysis reported. OC content of sediment ca 4%**		**2**	**Mayor *et al* 2008**

Crustaceans							
Corophium volutator	10 d	LC_{50}	193 (wwt)	Mean measured concentration	No details of sediment characteristics available	4	WRc (2000)
Corophium volutator	10 d	NOEC (mortality)	115	Mean measured concentration	No details of sediment characteristics available	4	WRc (2000)
Corophium volutator	10 d	MATC (mortality)	190	Mean measured concentration	No details of sediment characteristics available	4	WRc (2000)
Corophium volutator	10 d	LC_{50}	6.32	Test in absence of sediment, high control mortality. Mean measured concentration	Absence of sediment affects relevance of the study and may impact results observed	3	WRc (2000)
Corophium volutator	10 d	NOEC (mortality)	3.2	Test in absence of sediment, high control mortality. Mean measured concentration	Absence of sediment affects relevance of the study and may impact results observed	3	WRc (2000)
Corophium volutator	**10 d**	**LC_{50}**	**141.5 (dwt; 2211 normalised to 5% OC)**	**Static study. Test guideline OSPAR 2005 Part B. Mean measured concentrations. 0.32 % OC.**		**2**	**EPP (2018c)**
Corophium volutator	10 d	NOEC	99.4 (dwt; 1553 normalised to 5% OC)	Static study. Test guideline OSPAR 2005 Part B. Mean measured concentrations. 0.32 % OC.		2	EPP (2018c)
Corophium volutator	**10 d**	**LC_{50}**	**255 dwt; 319 normalised to 5% OC)**	**nominal concentrations,no analysis reported. OC content of sediment ca 4%**		**2**	**Mayor *et al* 2008**
Pandalus platyceros	8 d	LOEC (mortality, genetic changes)	42	Flow through, measured concentrations	Sediment OC <0.5%	2	Veldhoen *et al* (2012)
Pandalus platyceros	**8 d**	**EC_{20} (mortality, genetic changes)**	**400 (dwt)**	**Flow through, measured concentrations**	Sediment OC <0.5%	**2**	**Veldhoen *et al* (2012)**
Homarus americanus	**10 d**	**LC_{50}**	**250 (330 dwt)**	**spiked sediment, flow through overlying water, measured concentrations**	**Field collected sediment from a control site in Canada; moisture content**	**2**	**Daoud *et al* (2018)**

					ca 22%; no details on OC content		
Marine chronic							
Annelids							
Capitella capitata	21 d	NOEC (effect not reported)	460	No study details available		4	Schering-Plough Animal Health (2000) cited by Telfer *et al* (2006)
Hediste diversicolor	**28 d**	**NOEC (survival)**	**283 (dwt; 615.2 normalised to 5% OC)**	**Test guideline ASTM E1611-00, OC content 2.3%, mean measured test concentrations**	**No effects at highest test conc**	**1**	**Fox (2019)**
Hediste diversicolor	**28 d**	**NOEC (growth)**	**283 (dwt; 615.2 normalised to 5% OC)**	**Test guideline ASTM E1611-00, OC content 2.3%, mean measured test concentrations**	**No effects at highest test conc**	**1**	**Fox (2019)**
Nereis virens	**30 d**	**NOEC (growth rate)**	**ca. 240 (dwt)**	**spiked sand, flow through conditions. Measured concentrations.**	**only 1 test concentration. Statistically significant effect on growth rate, and behaviour (burrowing). Test medium silica sand overlayed with laboratory water (no OC added)**	**2**	**McBriarty *et al* (2018)**
Crustaceans							
Leptocheirus plumulosus	28 d	LC_{50}	49.7 (44.2 – 55.5) (dwt; 776.6 normalised to 5% OC)	Semi-static test. Test guideline EPA/600/R-01/020, measured concentrations, 0.32% OC		2	EPP (2018e)
Leptocheirus plumulosus	28 d	NOEC (mortality)	21.7 (dwt; 339.1 normalised to 5% OC)	Semi-static test. Test guideline EPA/600/R-01/020, measured concentrations, 0.32% OC		2	EPP (2018e)

Leptocheirus plumulosus	28 d	NOEC (growth rate; mean weight per surviving adult)	<21.7 (dwt; <339.1 normalised to 5% OC)	Semi-static test. Test guideline EPA/600/R-01/020, measured concentrations, 0.32% OC		2	EPP (2018e)
Leptocheirus plumulosus	**28 d**	**EC_{10} (growth rate; mean weight per surviving adult)**	**17.6 (dwt; 275 normalised to 5% OC)**	**Semi-static test. Test guideline EPA/600/R-01/020, measured concentrations, 0.32% OC**		**2**	**EPP (2018e)**
Leptocheirus plumulosus	28 d	LC_{50}	220 (180 – 260) (dwt; 3666.7 normalised to 5% OC)	Flow through test. Test guideline EPA/600/R-01/020, measured concentrations, 0.3% OC		2	EAG (2018)
Leptocheirus plumulosus	**28 d**	**EC_{10} (male growth rate)**	**57 (dwt; 950 normalised to 5% OC)**	**Flow through test. Test guideline EPA/600/R-01/020, measured concentrations, 0.3% OC**		**2**	**EAG (2018)**
Leptocheirus plumulosus	**28 d**	**EC_{10} (female growth rate)**	**49 (dwt; 816.7 normalised to 5% OC)**	**Flow through test. Test guideline EPA/600/R-01/020, measured concentrations, 0.3% OC**		**2**	**EAG (2018)**
Leptocheirus plumulosus	**28 d**	**EC_{10} (reproduction)**	**43 (dwt; 716.7 normalised to 5% OC)**	**Flow through test. Test guideline EPA/600/R-01/020, measured concentrations, 0.3% OC**		**2**	**EAG (2018)**
Corophium volutator	**28 d (&75 d)**	**NOEC (survival, growth, reproduction)**	**61.28 (dwdwt; 53.3 normalised to 5% OC)**	**Semi static test. Test method adapted from literature method. Measured concentrations, 5.75% OC**	**No effects observed at highest test conc**	**2**	**Scymaris (2018)**
Corophium volutator	28 d (& 75 d)	LOEC (survival, growth, reproduction)	>61.28 (dwt: >53.3 normalised to 5% OC)	Semi static test. Test method adapted from literature method. Measured concentrations, 5.75% OC	No effects observed at highest test conc	2	Scymaris (2018)

Homarus americanus	**30 d (extended to 71d)**	**NOEC (growth)**	**34 (45 dwt)**	**spiked sediment, flow through overlying water, moisture content 23 - 25%**	**interstage growth endpoint . Field collected sediment from a control site in Canada; moisture content ca 22%; no details on OC content**	**2**	**Daoud *et al* (2018)**
Homarus americanus	**30 d (extended to 71d)**	**NOEC (behaviour)**	**<8.8 (<11.6 dwt)**	**spiked sediment, flow through overlying water, moisture content 23 - 25%**	**behaviour (position on back) endpoint. Field collected sediment from a control site in Canada; moisture content ca 22%; no details on OC content**	**2**	**Daoud *et al* (2018)**

Table A1.3: microcosm data

Study	Species	Test duration	Endpoint	Result (µg/l)	Test method	Comment	Reliability	Reference
Freshwater								
Outdoor microcosm	phytoplankton, zooplankton and invertebrates	139 d	No Observed Ecologically Adverse Effect Concentration (NOEAEC)	3 * 0.3	Static, nominal concentrations		1	EC (2011)
Marine								
Outdoor microcosm	*Arenicola marina*	28 d	NOEC (growth, survival)		2% OC	No effects on species in test. However high variability in effects and mortality	3	Cheng *et al* 2020

						in controls mean results difficult to interpret.		
Outdoor microcosm	*Coropium volutator*	28 d	NOEC (growth)	30 (dw; 75 normalised to 5% OC)	2% OC	Although dose-response observed for reported LC50 of 316 µg/kg dwt, mortalities in controls of 41.3 and 31.7% mean results difficult to interpret. Authors do note on wwt basis LC50 result similar to other studies (190µg/kg wwt)	3	Cheng *et al* 2020

Outdoor microcosm	*Cerastoderma edule*	28 d	NOEC (growth, survival)		2% OC	No effects on species in test, however high control mortalities – 60 and 65%	3	Cheng *et al* 2020

Annex 2 Summary of 2019 UKTAG Consultation comments and responses

Thirteen responses were received on the proposal for a revised environmental quality standard (EQS). UKTAG asked two questions in relation to this standard; question 9 asked if stakeholders support the derivation of the proposed EQS and question 10 asked whether there is any other relevant data that has not been considered in the derivation of the EQS.

Of the responses received one fully agreed with the derivations and resulting EQS values. Other responses identified reasons why, out of the three EQS presented, they believed revision to the proposals for the water Maximum Acceptable Concentration (MAC) EQS and sediment EQS were required. Some respondents believed the proposal to be too stringent, others too permissive. Reasons for these views included: the choice of assessment factors used in the derivations; whether assessment factors used to derive the MAC were sufficiently protective of all aquatic species, including larval stages of commercially important species; the use of freshwater insect data in setting marine standards; and possible differences in sensitivities between marine and freshwater organisms (in relation to the proposed sediment standard). One response provided results of three additional long-term sediment toxicity studies. For one of these studies, a short summary report was also provided. These new data and information are potentially significant in terms of the derivation of a sediment EQS and will be considered alongside all the other comments and responses provided. We have not received any data in support of a more precautionary standard than the original recommendation from UKTAG of 23.5 ng/l.

We have summarised the remaining responses under appropriate headings. Full details of the comments received and responses are available on the UKTAG website.

Methodology – selected assessment factors

We received a number of comments on the assessment factors used in setting the pelagic MAC EQS and the sediment EQS. For the MAC EQS, we will reconsider the dataset alongside the comments raised re: protection of all aquatic species including larval stages of commercially important species and the assessment factor used. For the sediment EQS, we were made aware of significant new data. We will ask for study reports or robust study summaries to be made available so that we can review this additional data, which may lead to a revised EQS proposal including a change to the assessment factor applied.

Data Interpretation – use of Arenicola data (in sediment EQS)

We received a number of comments which are supportive of not using the sub-lethal endpoint from the acute Arenicola study in the derivation of the sediment EQS. Some responses also commented on the lack of a chronic study for this species and its relative sensitivity. A new study has been conducted for a ragworm species (one of the three referred to above). As part of our review of the new submitted data (assuming it is made available), we will consider its relevance to Arenicola.

Data interpretation – use of insect data (in sediment EQS setting)

We received a large number of detailed comments on the use of freshwater insect data in setting a marine EQS. The majority of these were not supportive because they believed insect species are less relevant for the marine environment being fairly rare and found only in intertidal zones. In addition, to date, the industry that uses the substance as the active ingredient in a veterinary medicine has been regulated only through surveys of impacts on subtidal benthic communities. In considering the comments received, we will seek further expert advice on the use of such species in the protection the marine environment. We will also seek policy advice on what the EQS for this substance is trying to achieve in relation to the protection goals of a marine EQS for a specific pollutant (which include all

areas within the marine environment from transitional and coastal waters up to three nautical miles off shore).

Data Interpretation – comparing fresh and marine water datasets; mode of action and statistical factors

We received a number of detailed comments on differences in sensitivities of fresh- and marine organisms to the chemical's mode of action, as well as a statistical demonstration including the three new sediment studies that the difference between the fresh and marine sediment datasets toxicities was statistically significant. The former will be considered as part of the work noted above to consider the use of a freshwater insect to derive a marine EQS. In terms of assessing whether the fresh and marine data are statistically different, we will look Page | 10 further at the complete datasets, including the new study data. Further comments on this aspect are included in the Annex's technical comments.

Data Interpretation – field studies

We received conflicting comments on the use of the field data in this derivation. The majority disputed the findings of the SEPA study, with one submission having apparently conducted reanalysis of the data. We will reconsider the two available field studies taking into consideration the comments received. This may include letting a contract to a third party to reanalyse all the data, provided all the required study details are made available to us.

New ecotoxicity test data

Several respondents referred to additional studies being available on the toxicity of emamectin benzoate to aquatic organisms. SSPO provided further detail on these additional data, which comprise:

1. Chronic 28-day growth study for the ragworm Hediste diversicolor;

2. Life cycle toxicity study for the sediment-dwelling midge Chironomus dilutus;

3. Life cycle toxicity for the amphipod Hyalella azteca.

We have requested further details of these studies so that we can verify their reliability for use in the derivation of the sediment EQS. These data greatly extend the available database for sediment toxicity and will be invaluable in the derivation.

Further to the comments received during the consultation, we will take the following actions:

- Request access to study reports or robust study summaries of the three new chronic toxicity tests in sediment dwelling organisms, and review their suitability for use in the derivation of an EQS for emamectin benzoate.

- Conduct further review of the two available field studies through an external independent third party.

- Consider further the protection of the marine environment and marine activities' regulation, in relation to the protection goals that exist for River Basin Specific Pollutants.

- Produce a revised EQS proposal based on consideration of the new studies, the further analysis of the field data and consideration of the comments received. This will be subject to independent peer review, either in full or targeted to its critical elements and reflective of comments received during the consultation.

· Forward our final recommendation to UK Administrations.

- As part of this process we will, as far as possible, make available relevant data.

Completion of the work outlined above is unlikely to be achievable before summer 2020 due to the number of steps and the need to involve external experts and organisations.

The proposed EQS will not be finalised until all relevant work identified above has been undertaken

Annex 3 Evaluation of field study reanalyses

A staticitcian was engaed to review the statistical approaches used in Bloodworth et al 2019 (based on the work reported in SEPA 2018) and reported additional investigations of the data collected for that and for the passive field monitoring study (SAMS 2018), repeating the analyses summarised in section 3.2.3 of this report but on five subsets of the data, and repeating the approached used in Bloodworth et al 2019 on the same five subsets and the SEPA 2018 study data in isolation (Dixon PM 2020b).

The author states that two aspects of Bloodworth et al 2019 approach seem unnecessary or contradictory. In part, this opinion seems to be based on a different statistical philosophy when it comes to attempting to solve or elucidate the complex problems that studies of this sort deal with. The author disagrees with the prescreening of variables using the variance inflation factor (VIF) to remove those that are moderately correlated, and the use of a likelihood ratio test (LRT) following Akaike information criterion (AIC) variable selection. In the author's opinion, VIF leads to early decisions to ignore certain variables that are moderately/highly correlated with other variables, although these do not cause problems for AIC-based model selection because AIC inherently avoids including such moderately/highly correlated variables in the same model. The author believes that using LRT after choosing models using AIC needlessly combines two statistical philosophies with different characteristics. The author goes on to state that regression coefficients ought, for risk analyses, to be for unstandardized coefficients rather than standardised coefficients as is the case in Bloodworth et al 2019. The authors of the Bloodworth et al 2019 study were given the chance to respond to these comments, and their response is summarised here (personal communication, 2020). Variables were pre-screened to look for relationships between explanatory variables, a common statistical practice although not universally practised. The Variance Inflation Factor (VIF) test was used to test for multicollinearity between the explanatory variables. (Variables that failed this test were removed from the analysis so that variance in the regression coefficients is reduced; this practice is well documented for the tests in this study (Zuur et al 2010)). In their statistical methodology, Bloodworth et al used the ΔAIC test to select the best fitting models from a global model containing all variables (Bolker et al., 2009). Where there was a simple nested model in the best fitting models from the ΔAIC test they used LRT to assess whether there were statistically significant differences between the most simple nested model and more complex models with more explanatory variables. If there was no statistically significant difference the simplest model was selected as the best fitting model, otherwise model averaging was used. Explanatory variables were mean centred so that their effects within the model were comparable; this is a suggested step for GLMMs as outlined in technical guidance such as Bolker et al (2009) and Zuur et al (2010). Bloodworth et al tried remodelling without standardising the input variables, and EMB still had the largest effect on crustacean abundance and richness, and was still statistically significant. They decided to use EMB residues in the set of variables submitted for model selection, ie to create a global model with all variables and then use the model selection process to select the best model. A different methodology may have been to consider all other parameters except EMB concentration to create a model and then add in EMB to a hypothetical prediction based on interquartile range values of other environmental predictors see if model fit was improved. Bloodworth et al did this as part of their analysis, and TOC came out as a significant predictor but with a much weaker effect. They recognised that there is some correlation between EMB and TOC, but state that the inclusion of EMB leads to a much stronger effect on crustacean abundance and richness than TOC (as described in Bloodworth et al., 2019). The authors also stated that sediment moisture content was removed from sediment characteristics prior to analysis because it failed the VIF test, but that including moisture content did not yield a better fitting model. They also state that the most important sediment characteristic, particle size, was not removed from any of the models.

Annex 4 Further information on Peer Review (for the current report)

Chemistry Task Team identified potential candidate peer reviewers, recognised as experts in their field, to act as independent peer reviewers of this report between September 2021 and November 2021. Two reviewers were engaged under contracts let by SEPA (as the UKTAG partner organisation funding the peer review). CTT set four specific questions for peer reviewers, in addition to the request to carry out a general review of the report. Peer reviewers were provided with a template to document their reviews and were asked to suggest specific actions to address their comments where relevant.

Specific Questions were:

1) Based on the use pattern and substance properties, have the correct compartments for QS derivation been identified?

2) Has the correct approach to data pooling been used (freshwater and marine data have been pooled)?

3) For the $QS_{sed,\ sw\ eco}$, data for a freshwater insect study are used to derive the QS. In setting a marine sediment QS for a river basin specific pollutant under the Water Framework Directive, is it appropriate to use insect data? If so, has the correct assessment factor been used, considering the available field study data and information on mode of action?

4) For the MAC-$QS_{sw,\ eco}$ and AA-$QS_{sw,\ eco}$, have the key data and assessment factors been chosen appropriately?

All comments submitted by the two peer reviewers were taken into account and acted upon in drafting this report. Specific comments and subsequent changes are identified in the relevant sections of the report.